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TROPICAL DISEASES BUREAU.

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PIROPLASMOSIS.

- (1) NUTTALL (G. H. F.) & STRICKLAND (C.). On the Occurrence of Two Species of Parasites in Equine "Piroplasmosis" or "Biliary Fever."—*Parasitology*. 1912. Feb. Vol 5. No. 1. pp. 65-96. With 1 plate, 1 text figure, and 5 charts.

According to these authors there are included under the term *Piroplasma equi* two separate and distinct parasites, and consequently two distinct diseases have hitherto been included under the name piroplasmosis.

In their view the true piroplasms always multiply in a characteristic manner, the peculiar piriform parasites developing by a process resembling budding from large rounded or slightly amoeboid parasites, the whole of whose protoplasm flows into the buds and gives rise to two piriform parasites without any residual body being left.

The description of the *Piroplasma equi* given by LAVERAN, who examined stained specimens sent by THEILER (1901), does not indicate this as the typical method of multiplication. FRANÇA convinced of the difference between Laveran's parasite and the *Piroplasma*, has placed the parasite in a new genus, named *Nuttallia*.

These parasites do not multiply according to the method described for *Piroplasma*; they do not occur as pairs of piriform parasites, and they form distinctive "cross-forms" which BOWHILL and FRANÇA regard as multiplication forms.

The occurrence of two types of parasites in equine piroplasmosis was pointed out by KOCH (1905), who believed that they might cause distinct diseases.

A short preliminary note was published upon this subject by the present authors in 1910, in which they showed that two distinct parasites occur in horses suffering from biliary fever, and consequently two distinct diseases are included under the name.

A strain of the *N. equi* was obtained from MCFADYEAN, the original having been obtained from South Africa and the strain maintained by passage through a number of horses. The blood was taken from a horse that had been inoculated with positive results two years previously.

To avoid confusion the authors first describe their observations on the *N. equi*, subsequently dealing with the true *Piroplasma* (*P. caballi* Nuttall 1910), which also occurs in horses suffering from biliary fever.

Nuttallia Equi.

Horse 1.—Inoculated with blood obtained from McFADYNEAN. Parasites appeared on the 8th day and the horse died on the 11th day.

Horse 2.—Inoculated with blood from Horse 1 on the day of its death. Parasites appeared on the 7th day and disappeared on the 22nd day. Horse recovered and still alive (after more than 2 years).

Horse 3.—Inoculated with blood from Horse 2 about seven months after the disappearance of the parasites. Parasites appeared on the 10th day and the horse died on the 20th day.

The living parasite.—This was studied in fresh blood films at body temperature.

The number of infected corpuscles was small. In Horse 1 it was 5.4 per cent.; in Horse 3, 13.2 per cent. In Horse 2 the parasites appeared in the blood in such small numbers that there was no adequate opportunity of counting them. The following types of parasites were studied:—

Small parasites.—Measured 1.1-1.4 microns, frequently amoeboid, moving about within the corpuscle while altering their shape. Occasionally, filiform or bud-like processes were protruded and retracted. The escape from corpuscles was never observed, nor were any of the small parasites observed to divide.

Single medium-sized parasites behaved in much the same way.

Very large single parasites, of which a certain number may be found, may show very little change of form, but they may assume an elongated or plump piriform shape.

Dividing forms.—In a number of cases stages of development of a single parasite into four were observed. In one case a previously rounded parasite was observed to give rise to four parasites after developing into a cross-form. In the majority of cases the cross-forms break up into four distinct parasites which wander away from each other, but in a very small number of cases five and six small parasites were observed to result from the breaking-up of a cross-form. The resulting young parasites are very active.

In a few cases multiplication occurred by a process of budding, but such appearances may be deceptive, for when an infected corpuscle ruptures, the main mass of the protoplasm may be seen to be connected with the supposed daughter individuals by delicate strands of protoplasm.

The escape of the young parasites resulting from the breaking-up of cross-forms was observed to occur in some instances soon after their separation from each other. The corpuscle was not ruptured in the process. The escaped parasites after swimming about vigorously for a few minutes disappeared, the conditions *in vitro* being doubtless unfavourable to their continued existence.

Although the authors are convinced that the young parasites under natural conditions immediately enter a new corpuscle, such

an entry was not actually observed. On one occasion a medium-sized parasite was observed to leave a corpuscle and degenerate without injuring the corpuscle. It was noted several times that the first parasites to escape were the small ones resulting from the breaking-up of cross-forms. Some of the corpuscles vanished while the contained parasite remained motionless, and in other cases the corpuscles burst suddenly and the parasites were ejected into the plasma.

Stained parasites.—The parasites stain in a manner similar to *Piroplasma*. About 90 per cent. of the invaded corpuscles contained single parasites of various shapes and sizes. About 2.5 per cent. contained 2-4, and 1.5 per cent. dividing or cross-forms. The number of free parasites encountered differs widely. In Horse 3 only a very small percentage of free forms were found (0.2-6 per cent.) and in Horse 1 as many as 26 per cent.

A diagram is given showing the "usual mode of multiplication of the *Nuttallia equi* in the circulating blood." The minute piriform parasite enters a fresh corpuscle and increases in size, is slightly amoeboid, with a general tendency to assume a pear-shape. When the parasite has attained a certain size, definitely amoeboid movements are observed. Judging from the form of the chromatin in stained specimens the next steps are as follows. The chromatin changes its shape from round to elongated and lies at the periphery of the parasite in the form of a curved rod. This then assumes a V-shape, small thickenings appearing at the free and the united ends of the arms of the V. The arms themselves disappear leaving the knob-like thickenings only, which become separated at equal distances from each other. The cytoplasm then shows a tendency to be pinched off into the form of a cross, one of the chromatin masses being in each arm. These stages, from the separation of the four little chromatin masses to the formation of the cross, have been observed in moist preparations.

Distribution of the parasites in the body.—The parasites appeared to be fairly uniformly distributed in all the organs examined.

In the case of Horse 3, which had been previously infected with *P. caballi* (see later), there were roundly 9 million red corpuscles per c.mm. at the time when parasites appeared, and by the 19th day this number had fallen to 3 million. Differential counts were made on four days, and it was found that there was a marked decrease in the number of eosinophiles.

Piroplasma (Babesia) caballi.

Horse 1.—Inoculated with blood obtained from MARZINOWSKY. Parasites appeared on the 15th day. Horse died on the 22nd day.

Horse 2.—Inoculated from Horse 1. Parasites appeared on the 8th day. Horse recovered. This animal was subsequently inoculated with the *N. equi* and is No. 3 of that series.

Horse 3.—Parasites appeared on the 9th day. Horse died on the 19th.

The living parasite.—Although the scarcity of the parasites in the blood made it impossible to make a complete study of the

development *in vivo*, the authors are convinced that *P. caballi* develops in a manner similar to the *P. bovis* and *P. canis*.

The forms of parasites seen correspond exactly with those observed in *P. bovis* and *P. canis*, and the process is slower than in the case of the latter.

In stained preparations the parasite appears to be very closely allied to *P. bovis*.

The predominant forms found in stained preparations were the rounded and double piriforms.

In Horse 3 there was a relatively slight decrease in the number of red corpuscles and a slight fall in the amount of haemoglobin. The leucocytes showed a decided decrease as the disease advanced, accompanied by a disappearance of eosinophiles and an increase in neutrophiles.

Immunity test with regard to N. equi and P. caballi.—As already mentioned Horse 3 of the *N. equi* series is the same animal as Horse 2 of the *P. caballi* series.

It was first inoculated with *P. caballi*, suffered from a mild infection, and to all appearances made a good recovery.

Two months later it was inoculated with *N. equi* and died of "biliary fever" 20 days later. Only *N. equi* could be found in the blood. This was the only horse of the six that had haemoglobinuria, and it is possible that the mixed infection was responsible for this.

Conclusions.—

The term "Biliary Fever" or "Piroplasmosis," hitherto supposed to apply to a specific disease affecting horses, in reality refers to two distinct diseases produced by distinct parasites. For convenience' sake, and in accordance with the terminology at present in vogue, these two diseases may be named after the parasites which produce them, *i.e.*, Piroplasmosis (due to *Piroplasma* [or *Babesia*] *caballi* Nuttall, 1910) and Nuttalliosis (due to *Nuttallia equi* (Laveran, 1910, França, 1909).

- (2) SCHUBERG & REICHENOW. Über Bau und Vermehrung von *Babesia canis* im Blute des Hundes. [The Morphology and Multiplication of *Babesia canis* in the Blood of the Dog.] *Arbeit. a. d. Kais. Gesundheitsamt.* 1912. March. Vol. 38. No. 4. pp. 415-434.

The authors employed principally Schaudinn's method of moist fixation followed by staining with Giemsa's solution. Examinations were also made of the living parasite on the warm stage.

The strain used at the commencement of the experiments was a very virulent one, young dogs dying in 3-4 days after infection. Owing to circumstances the experiments had to be interrupted and it was found that the strain in the infected dogs had lost much of its virulence and that it was very difficult to re-instate this.

The small number of parasites present in the blood of some of the dogs enabled the authors to investigate their distribution in the body. Large masses of parasites were found in the kidneys and also in the capillaries of the lungs. There is a striking difference between the number of parasites in the peripheral capillaries and the large vessels, and the authors state that, if at

the onset of the temperature reaction a small cut be made in the ear, the first drop of blood which escapes may be very rich in parasites, while the second may be so poor that prolonged search is required to find even single ones.

In fresh preparations of such blood there may frequently be observed masses of blood corpuscles which are adherent to each other. The majority of such corpuscles may be invaded with *Babesia*. This is explained by the fact that in cases of piroplasmosis the corpuscles have a tendency to cling together, and that this causes plugging of the capillaries and the multiplying parasites naturally invade the cells nearest at hand.

Agglutination of red cells begins before the parasites make their appearance in the circulation, and it may occur as early as 48 hours before the blood invasion. The authors found in one case at least that although there was obvious agglutination of the red corpuscles in blood taken from the ear, it was not discoverable in the heart blood when the animal was killed soon after.

Canine piroplasmosis is associated with a marked increase in the number of mononuclear leucocytes, and these frequently contain not free parasites, but entire invaded red corpuscles.

Morphological changes observed during multiplication.—The authors agree with NUTTALL and GRAHAM SMITH that the pairs of pear-shaped parasites are formed by a process of budding from rounded parasites, the buds growing at the expense of the mother-cell. They believe however that the rounded cell which gives rise to the daughter cells is not intra-corpuscular. They hold the view that the amoeboid cells which precede the rounded parasites exhibit movements that are too active to permit of the view that the parasites are intra-corpuscular. Comparison is drawn between these and the slow movements exhibited by the merozoites of tertian malaria which are intra-corpuscular.

The parasite regains the interior of the red corpuscle according to the authors by means of the buds which develop into the pear-shaped parasites. These are described as boring into the corpuscle and there developing at the expense of the mother cell outside. The parasites present in multiple infection of a red cell are almost always developed from a single parasite as is evidenced by the fact that the parasites are all at the same stage; the authors have however occasionally observed multiple infection with the parasites in different stages. The pair of parasites developed from the first amoeboid parasite again pass to the outside of the cell, become amoeboid, and penetrating again by means of their processes double their infection, and so on. The authors have never observed more than 16 parasites in a single corpuscle. They have, further, never observed the simultaneous formation of four pear-shaped parasites as described by NUTTALL and SMITH, and CHRISTOPHERS.

Cytology.—The cytoplasm of the *Babesia* is finely alveolar in structure. In practically every parasite one or two clearer portions can be seen. In dry preparations these appear like vacuoles, but in preparations that have been fixed moist fine strands of protoplasm are observed to traverse the apparent spaces. These "vacuoles" are certainly not part of the nuclear structure, since in many parasites the nuclear substance is quite outside them.

In the majority of the pear-shaped and amoeboid parasites the nucleus is in the form of a single, round, homogeneous mass of chromatin which possibly contains a minute paler portion. When an amoeboid parasite is about to divide the mass of chromatin becomes elongated, one pole appearing to be richer in chromatin than the other. The two poles then separate from each other and finally two rounded masses of chromatin are formed. Each again divides and while this is in progress the buds begin to form, one portion of the chromatin going into each bud and then a second portion. In this way the two pear-shaped parasites are each provided with two pieces of chromatin. The difference in texture between the two pieces is now very obvious, one being large and the other paler and smaller. When the twin parasites separate this appearance of two portions of chromatin is lost. Delafield's haematoxylin is superior to Giemsa's stain for showing the changes in the chromatin.

The pear-shaped parasites which have been set free by the disintegration of their host cell appear in fixed preparations to be shorter than the intra-corpuscular forms. They also appear to be more intensely stained; a possible explanation of this being that the intracellular forms are flattened and consequently thinner. In the majority of cases the free parasites contain only one piece of chromatin.

The authors do not agree with the statements made by other investigators regarding the occurrence of flagellated forms.

Brief criticisms are given of the descriptions by various authors of the multiplication of the parasite in culture, and of the developmental stages in the tick, but nothing new is added.

The authors do not consider the second mass of chromatin to be representative of a blepharoplast but rather of a nucleolus, and they think that in the present state of knowledge *Babesia* should be considered to be more closely allied to the *Coccidia* than to the *Binucleata* (HARTMANN).

- (3) CIUCA (A.) Recherches sur l'Influence de la Splénectomie totale sur l'Évolution de la Piroplasmose canine. [The Effect of Total Splenectomy upon the Evolution of Canine Piroplasmosis.]—*Bull. Soc. Path. Exot.*, 1912. Feb. Vol. 5. No. 2, pp. 143-150.

After a brief reference to the varying opinions held by different authors regarding the effect of splenectomy in different diseases the author describes his experiments in connection with canine piroplasmosis.

His experiments were grouped into three classes:—

A. Those in which the spleen was removed from normal dogs which were subsequently infected.

B. Those in which the dogs were infected before excision of the spleen.

C. Those in which the spleen was excised when the piroplasmosis was at its height.

The conclusions drawn are as follows:—

1. Infection subsequent to removal of the spleen, provided the dog has completely recovered from the operation, does not affect the course of the disease, setting aside complications.

2. Splenectomy during the evolution of the disease undoubtedly aggravates the symptoms, and especially in young animals. This is the more evident in cases in which there is some debilitating condition as a complication of the disorder caused by the splenectomy.

3. The reappearance of the parasites and the aggravation of the symptoms are less marked in dogs that are operated upon from 3-12 days after the disappearance from the blood of the last infected corpuscles.

4. When the animal has passed through an attack and is on the road to recovery splenectomy in the great majority of cases does not lead to any reappearance of the parasites. In exceptional cases there is a slight and transient reappearance.

5. In dogs that are operated upon immediately after the disappearance of the parasites, and in those operated upon during the course of the disease there is often an immediate and very marked hyperleucocytosis, which steadily increases up to the death of the animal.

In all the experiments, and especially in those in which the animals were infected after splenectomy, one must hesitate before accounting for the conditions observed by the absence of the spleen. Operative shock and accidental infections appear to play a more important part than the absence of the spleen.

A practical conclusion may be drawn that splenectomy is not a practicable method of preserving the virus, and all the more in that even in animals that have not been operated upon the blood is always infective for some months and even a year without any parasites being visible in it.

- (4) INCHIOSTRI (H.). Vorkommen und Formen der *Piroplasma ovis* in Dalmatien. [Piroplasmosis of the Sheep in Dalmatia.]—*Oesterreichische Wochenschr. f. Tierheilkunde*. 1912. Vol. 37. No. 29, pp. 289-292; No. 30, pp. 299-302; No. 31, pp. 310-313; No. 32, pp. 320-322; No. 33, pp. 331-332; No. 34, pp. 340-343.

The author's investigations were carried out in the Zara District of Dalmatia. The typical piroplasms were found in smears of peripheral blood stained with various modifications of the Romanowsky method. Multiple infection of a single corpuscle occurs, as many as 8 parasites having been observed. Exact methods of measuring the parasite were not available, but taking the size of a red corpuscle as 4-6 microns the author judged that the parasites vary in size from $\frac{1}{2}$ -3 microns. Both intra- and extracorpuseular forms were encountered. During life the parasites are more numerous in the peripheral blood than in the large vessels. Piroplasms occur in large numbers in the spleen.

In moist preparations the smallest of the parasites and those that are undergoing division show very active movements. As a rule, the intracellular parasites remain in the same corpuscle until it ruptures, but occasionally piroplasms have been observed to leave corpuscles without causing their destruction. Such liberated parasites actively seek out another red cell, and penetrate it by

means of a pointed projection which is suddenly formed. Repeated attempts to penetrate different corpuscles have been observed.

Parasites have been observed in the interior of leucocytes as a result of phagocytosis.

In moist and stained preparations large round parasites have been observed which are provided with a long pseudopodium. Parasites of this type are rarely encountered within red cells, but are as a rule extraglobular.

The *Rhipicephalus bursa* is the sheep tick that occurs in the district, and in blood in these ticks the author has found the star-shaped parasites described by DUCHONKOWSKY and LUNS. In addition to these he has observed immense numbers of small rounded parasites which execute amoeboid movements. Only ring- and star-shaped parasites were observed in the fully engorged females, the small round forms occurring exclusively in the larvae and nymphs.

The predominance of pear-shaped parasites in stained preparations may be explained by supposing that contraction has occurred during the process of fixation.

The author found that the parasites in the spleen had lost all power of movement within 12 hours of the death of the host, but by the addition of salt solution the motility was restored and the parasites made to retain their vitality for a long time. Parasites are scarcely recognisable in the internal organs within 24 hours after death, and shortly after this they have entirely disappeared.

Parasites present in the blood have been proved to be living 8-10 days after the death of the host.

The author describes the occurrence of peracute, acute, abortive, and chronic cases of the disease, and gives a detailed account of the symptoms observed in each variety.

The peracute form occurs principally during the hot season of the year and almost exclusively attacks ewes that are in poor condition. The temperature rises suddenly and may reach 42° C. or more. Before death, which occurs very shortly, the temperature falls again, and is almost invariably subnormal at the time of death.

Jaundice is a very constant symptom. There are frequently observed symptoms of colic, and rigors are prominent. There is always haemoglobinuria in the peracute cases.

The following lesions are found: Haemorrhages and gelatinous infiltration in the connective tissues; petechiae in the muscular masses; jaundice; hyperaemia and oedema of the brain with haemorrhages in the dura and spinal membranes; swelling and infiltration of the red marrow of the long bones. There is always enlargement of the spleen, but not to the extent observed in anthrax. The spleen pulp is never fluid and the author has observed only one case of rupture of that organ.

In the acute form of the disease the duration of illness is longer, the temperature does not rise so high and some cases recover. Haemoglobinuria does not persist for very long; in many cases it ceases after a few hours. Frequently there is blood-stained diarrhoea.

Acute cases last from 3-5 days, and in those in which the animals are going to recover the acute symptoms abate in 10-12 days. Convalescence is very slow. There is often complete loss of fleece in animals that survive an acute attack. Relapses are not uncommon.

A mild form of the disease occurs in the spring and autumn. It is rarely seen in adult animals, but principally affects the lambs.

An experiment was carried out with the object of testing the difference in the resistance offered to the disease by strong and by weakly animals, the inoculation material being virulent blood and blood from a recovered animal. The resistance offered by indigenous sheep as against that offered by other sheep was also tested.

Five animals in good condition and five weakly ones were inoculated with blood from an immune animal, with the result that all the five healthy animals recovered from the disease so induced and two of the weakly animals died. A test carried out with fully virulent blood (blood containing numerous parasites) had similar results.

Three sheep from Croatia inoculated with immune blood all succumbed.

In order to ascertain for what period animals remained infective after an attack of the disease, a number of animals were inoculated with blood from immune animals at intervals varying from 2-11½ months after the recovery of the immune animals from the attack which conferred immunity upon them. The dose in each case was 10 cc.

Only those inoculated at an interval of 2 months died. Those inoculated at 3½ and 5 months suffered from a mild attack but recovered, and those inoculated subsequently did not react. The blood used for those animals that died contained piroplasms, but in that used for subsequent animals none could be found. The author has been able to demonstrate piroplasms in the blood for periods varying from 2 to 12 months after recovery. Those cases in which the parasites persist for a long time he terms "latent" cases.

Chronic cases are those in which there are frequent slight relapses, and the course of such cases closely resembles that followed by cases of infection with liver fluke, the affected animals gradually wasting away and becoming generally cachectic.

A number of drugs were tried by the author in the treatment of the disease, but none gave constantly satisfactory results.

ANAPLASMOSIS.

- (5) THEILER (A.). Weitere Untersuchungen über die Anaplasmosis der Rinder und deren Schutzimpfung. [Further Investigations regarding Anaplasmosis and Protective Inoculation against the Disease.]—*Zeitschr. f. Infektionskrankh., parasit. Krankh., u. Hyg. d. Haust.* 1912. March 27. Vol. 11. Nos. 3-4, pp. 193-207.

Having given a brief resumé of his reasons for believing *Anaplasma* to be a distinct species of parasite, the author proceeds to describe experiments in support of this view.

1. The Separation of Anaplasmosis from Babesiosis by proper Selection of Ticks.—

As is well known an animal that has passed through an attack of either Texas fever or anaplasmosis, although immune, is a carrier of parasites, which may be transmitted to susceptible animals either by ticks or by inoculation with blood. If an imported English animal be inoculated with blood from an animal that has been bred in a tick infested district, one or other, or as a rule both, of the diseases may be expected to develop. For a number of years the author has observed that calves which he has obtained from the Karoo, which is only lightly infested with certain species of ticks, have shewn a striking degree of susceptibility to redwater and only a slight degree to anaplasmosis. This aroused the suspicion that these calves were possibly immune to anaplasmosis, the animals having passed through an attack when quite young, and that the animals acted as carriers of the parasite.

This view received some support from the fact that the blue tick (*Rhipicephalus decoloratus*) was unknown in the district, the black-pitted tick (*R. simus*), which together with the blue tick is a host of the *Anaplasma*, alone being found. Calves were obtained from two Karoo farms. Experiments were made on calves a few weeks old born on the premises of the laboratory, in order to ascertain whether the *Babesia bigeminum* could really be excluded.

Three young calves were inoculated with blood from the same number of Karoo calves with the result that two shewed a double infection and the third a pure anaplasma infection. The Karoo calf from which the anaplasma (pure) infection was derived was obtained from a different farm to the others. The experimental calf was subsequently inoculated with immune babesia blood and developed a typical attack of Texas fever, the parasites being fairly numerous in the blood. This calf was used as the starting point of a series of inoculations into susceptible English animals.

Twelve English animals and one Africander calf were inoculated, the dose varying from 1-100 cc. In no single case was there any infection with the *Babesia bigeminum*, shewing that a pure infection with the *Anaplasma* occurs naturally.

Some of these animals were afterwards tested with a view to ascertaining whether they were immune to redwater. Six were inoculated with blood, and three by means of ticks. With one exception, in which case the reaction was of a doubtful nature, all the animals reacted typically, and in some instances the reactions were so severe that treatment with trypanblue had to be resorted to.

A striking feature of the anaplasmosis infections was that none of the animals died, and that the disease ran a mild course in practically every case. The majority of the cases would have escaped observation if the thermometer and the microscope had not been used. This result was in striking contrast to results obtained with English animals in earlier experiments, and indicated a great variation of virulence. On microscopic examination it was found that the *Anaplasma* used in these experiments possessed certain characters by which it could be distinguished from the *Anaplasma* previously found.

Whereas the parasite first found was in the great majority of cases disposed towards the rim of the invaded red cell, and only exceptionally in the centre, the parasite observed in the series of experiments under consideration was most frequently found near the centre of the invaded blood corpuscle.

Apart from this there was a difference in size between the parasites. The difference could not be expressed exactly in figures, but was quite apparent to the skilled eye; those disposed at the margin of the corpuscles being generally larger than those at the centre. On these grounds the author concludes that there are two varieties of *Anaplasma*, and designates the form that is found centrally placed in the corpuscle as *Anaplasma marginale* (var. *centrale*).

2. Testing of Animals immune to *Anaplasma centrale* with *Anaplasma marginale*.—

Six animals that had recovered from red water and central anaplasmosis were inoculated with the *A. marginale* with the result that five reacted after the usual period, although the attacks were mild.

This shews that *A. centrale* protects against the *A. marginale* to the extent of preventing a severe attack of the disease, but the immunity is not complete. From a theoretical point of view therefore, the parasites, which are distinguishable from their position in the corpuscles and their difference in virulence, should be considered as varieties of one and the same species.

Notice should be taken of the fact that every animal whether suffering from Texas fever or anaplasmosis developed miltans piroplasmosis when injected with the *Babesia mutans*. This is further evidence of the specific nature of this parasite.

3. Protective Inoculation against Anaplasmosis.—

It having been shewn that the *Anaplasma marginale* var. *centrale* was the cause of a mild illness from which all the experimental animals recovered, and that such animals possessed a considerable degree of immunity against the *A. marginale* infection, the question naturally arose as to whether the *A. centrale* could be used as a protective inoculation against natural infection. Owing to the susceptibility of imported animals to the two diseases—redwater and gall-sickness—the question had to be decided whether the animals should first be immunised against redwater, controlling the inoculation with trypanblue if necessary, and then against the *Anaplasma*; whether the protective inoculations should be carried out in the reverse order; or whether the two should be carried out together.

(a) Inoculation against redwater followed immediately by inoculation against anaplasmosis.

Three animals were inoculated with anaplasma blood after the subsidence of a reaction due to previous inoculation with redwater. In all three cases reactions occurred at the end of the fourth week and were very mild, the maximum percentage of invaded blood corpuscles being 8.6.

(b) Inoculation with *Anaplasma* followed after the subsidence of the reaction by inoculation with redwater.

Three animals were also used for this experiment. In each case the attack of anaplasmosis was slight, but the redwater caused by the subsequent inoculation with redwater blood on the 40th day was in one case so severe that resort was had to trypanblue; the other reactions were mild.

Three further animals were used for an experiment of the same nature. In the first animal there was a slight anaplasmosis reaction and redwater blood was inoculated on the 40th day. The resulting reaction was so severe that trypanblue was used. The two other animals developed very mild attacks of anaplasmosis but failed to react to the redwater inoculation, a *Babesia bigemina* infection having developed in both the animals during the incubation period of the anaplasma infection. This anomalous result might be explained either by the animals injected having contracted redwater infection naturally on the veldt, the anaplasmosis inoculation causing a relapse, or the animal from which the anaplasmosis infection was derived might in some way have become infected with the *Babesia bigemina*. The striking fact remains that the whole of the animals reacted. The second suggested explanation appears to be the more probable in view of the fact that the animal which supplied the blood for the anaplasmosis inoculations was found to be refractory to a redwater inoculation.

(c) Simultaneous inoculation with redwater and anaplasmosis.

Five animals received simultaneously doses of blood containing the *A. centrale* and *B. bigemina*, the doses varying from 5-50 cc. The results shewed that the procedure was without danger provided that the *Babesia* infection was controlled by the use of trypanblue. From a practical point of view an answer was required to a further question, whether animals so immunised could be immediately exposed to natural infection.

In view of the shorter periods of incubation in experimental cases than in natural cases of infection one might suppose that animals protected experimentally would resist natural infection.

Animals that had been inoculated with African redwater in England were inoculated with anaplasma immune blood after their arrival in Africa, and had placed on them while in the stalls a large number of infected ticks. After the subsidence of the reaction they were turned out. The object of placing the ticks upon them was to set up a natural attack within the period of incubation.

Five animals were used and in none of them was there any reaction that was clinically recognisable. Possibly the animals reacted to the *Anaplasma* after a long period of incubation, but as no clinical symptoms were observed further blood examinations were not carried out. Cattle that are immune to redwater may be exposed to natural infection with *Babesia bigemina* during the process of immunisation against *Anaplasma* without running any risk.

Conclusions.—At least two forms of anaplasmosis can be distinguished—a virulent and a less virulent. The diseases can be transmitted by inoculation with immune blood without any exaltation of virulence. Recovery from the mild form of the disease

protects against the severe form, but there is no complete immunity. The two diseases are recognisable microscopically from the size and position of the causal parasites in the red corpuscles. In the severe form the parasite is generally disposed towards the rim of the invaded cell (*Anaplasma marginale*) and in the mild form towards the centre (*Anaplasma marginale* var. *centrale*). The peripherally placed parasites appear to be larger than those that are centrally placed. This fact is of importance in connection with protective inoculation, and the results obtained in the field confirm the accuracy of this conclusion. As far as South Africa and indeed the greater part of Africa is concerned, protection must be conferred upon imported animals from Europe if the trade is to flourish. This is without danger if pure *Babesia bigemina* be used for the inoculation; that is to say, the virulent *Anaplasma marginale* must not be injected.

By means of methodical inoculations this pure strain can be maintained, as can a pure strain of the *Anaplasma centrale*. In the Laboratories of the Union of South Africa at Onderstepoort and Grahamstown both these strains were maintained in a state of purity.

(6) BASILE (C.). Sull'*Anaplasma canis*. [Canine Anaplasmosis].—*Pathologica*. 1912. June 15. Vol. 4. No. 87. pp. 358-360.

During the course of his examinations of the blood of a number of different species of animals in the country around Messina, the author encountered a parasite in the blood of a dog which was very like *Anaplasma*. The parasite was observed in smears made from the peripheral blood of an adult dog which was in a very anaemic and poor condition. The smears were fixed and stained with Giemsa.

An attempt was made to transmit the disease experimentally. With this object blood was drawn with aseptic precautions from the liver of the infected dog and injected intraperitoneally into a puppy about a month old. This animal was protected from blood-sucking insects, and its blood was frequently examined for parasites before the experiment was commenced without any being found. Another puppy of the same litter was kept as control, and its blood was examined for parasites with negative results.

A few days later the infected dog was killed in order to study the distribution of the organism in its body. The parasites were found to be very scanty in the spleen, scanty in the liver and numerous in the lungs.

The inoculated dog appeared to be perfectly healthy until the tenth day after inoculation. *Anaplasma* was found in smears of the hepatic and peripheral blood stained with Giemsa.

Before the experimentally infected dog was killed blood was drawn from the liver with aseptic precautions and injected immediately into the peritoneal cavity of a second puppy, another of the same litter being kept as control. The blood of this dog was also examined, but no parasites were found.

At the post-mortem of the first experimentally infected dog parasites were found to be scanty in the spleen and numerous in the liver.

The second experimental dog refused all food after 24 hours and died on the third day. The course of the disease was very acute, and parasites were found in the blood of the peripheral vessels, spleen and liver.

The control dogs were killed when they were in perfect health and no abnormalities were found at the post-mortem.

Morphology of the Parasite.—The author has observed two forms of the parasite: one a coccus-like body which is apparently devoid of cytoplasm, and the other a crescent-shaped body which possesses cytoplasm.

The coccus-like bodies are rounded or oval in shape and measure about 2 microns. They are found in smears of blood from peripheral vessels, liver, spleen and lungs. They may be free or enclosed in corpuscles. The intracorpuseular forms are either centrally or eccentrically placed. Double, treble, and quadruple forms have been observed, and the author believes this multiplicity stands in some relation to the method of multiplication of the parasite.

The diplococcus form sometimes presents a dumb-bell-like appearance. In preparations that are not intensely stained it is possible to distinguish a marginal portion stained an intense rose-violet surrounded by a clear halo. In the interior of the parasite may be observed a number of granules of chromatin more intensely stained.

The crescent form of the parasite was found only in the dog naturally infected and in the second of the experimental animals.

The semilunar bodies vary from 3.5-5.5 microns in length. The ends may be either rounded or pointed. They consist of a body and nucleus, the body being surrounded by a peripheral zone in the form of a membrane.

The peripheral zone is not constant, but when present it stains a pale pink, and in the author's preparations was only to be seen in those parasites in which the body was stained pale blue and had a granular appearance.

The nucleus, which measures from 1.2 microns, is either central or a little towards one end of the parasite. It sometimes shews a more or less centrally-placed vacuole. In some of the parasites the nucleus is elongated and in some it is double. Within the nucleus there is a granule of chromatin that is more intensely stained, and in some cases there can be seen another granule of chromatin either close to or a little removed from the nucleus.

In a preparation from the hepatic blood of the naturally infected dog the author observed a single parasite which appeared to be provided with a flagellum about 5 microns in length with a thickening at its free end. The body of this parasite measured 4.9 by 2.3 microns. The cytoplasm was vacuolated and contained a large nucleus.

The author considers it premature to attempt to give any definite interpretation of the forms of the parasite found.

TRYPANOSOMIASIS.

- (7) LEESE (A. S.). Biting Flies and Surra.—*Jl. Trop. Veter. Science.* 1912. Jan. Vol. 7. No. 1, pp. 19-32.

The author's object is to show that the doubts cast on the theory of the mechanical transmission of surra, in view of KLEINE's experiments with the *T. brucei* and the *G. palpalis*, are not justified. In support of his view that the *T. evansi* is commonly transmitted mechanically the author gives accounts of outbreaks of surra occurring in desert places where *Tabanus* is unknown.

In the monsoon season of 1908 and 1909, when the rainfall was excessive, there was an outbreak of surra in the Imperial Service Camel Corps. There were in all about 500 camels, and by January 1910 more than half of these were dead, and more than half of the remainder were affected. On tracing the history of these animals it was found that none of them had been out of the State (Bikanir) for duty and that only 40 of them had grazed outside the State in the monsoon season. All the animals save five had been sufficiently long in the service to prove that they could not have been affected at the time of purchase. The five were almost certainly diseased at the time of purchase. This obviated all difficulty of accounting for the source of the disease.

The author was able to prove that the outbreak occurred when the animals were grazing on a desert portion of the State at least 100 miles from the fly zones. Further, the State breeding herds and the animals belonging to the villagers also suffered, there being among the latter young animals that had never been beyond their village allotment. The only biting fly found was the *Lyperosia minuta*.

The second outbreak occurred among the camels kept for civil purposes in the same State during the monsoon season of 1910. The author had examined these animals in the previous January and had found them all healthy. The total number of camels was 44 and by the end of November 11 had died, and of the remainder 16 were affected. These animals had been kept together and had not been elsewhere than in or near Bikanir City and in Gajner, where there is an isolated garden belonging to the State with a tank in it, and in the desert between these two places, a distance of 20 miles.

Investigation showed that *Tabanus* is not found in these places, but that *Stomoxys* is present in both, and *Lyperosia* is very abundant; mosquitoes also occur in both places.

The inference drawn by the author from the facts brought to light by his investigation is that in these cases the *Lyperosia* was the principal agent involved in the spread of the disease.

This *Lyperosia* occurred in swarms on every camel, and set up intense irritation causing the animals to rub against each other. Possibly inoculation sometimes occurs in this way. The flies show no great tendency to pass quickly from one animal to another, but this must often take place as they are found in such enormous numbers and the hosts in these cases were grazed close together.

It was impossible to carry out experiments in connection with the mechanical transmission of the disease by *Lyperosia* during 1911 owing to the drought.

The presence of this fly is not essential to the spread of surra. In Sohawa, where equine surra was previously unknown, a single case occurred in a mare that stood about 25 yards from the camel camp. *Tabanus*, *Stomoxys* and mosquitoes were all found in this district. The author draws the following conclusions from his investigations:—

1. That surra can spread widely in the absence of *Tabanidae* (but in the presence of *Lyperosia*).

2. That surra can spread easily in the absence of *Stomoxys* and mosquitoes (but in the presence of *Lyperosia*).

3. That surra can spread in the absence of *Lyperosia* (but in the presence of *Tabanids*, *Stomoxys* and Mosquitoes).

4. That if *T. evansi* undergoes any life cycle in any of these biting flies, then either surra can spread so easily in the absence of the specific fly, if there are enough biting flies of other genera to do it mechanically, that the question of life-cycle loses its practical importance, or, that *T. evansi* is capable of developing in the bodies of several distinct genera of biting fly. Is it not far more likely that the common power of transmission is a mechanical one?

The author then criticises briefly the writings of a number of authors regarding the method of transmission of surra by the agency of flies and adds some notes on the occurrence of biting flies in the North-Western camel country of India.

All the biting flies except the *Hippoboscids* require moisture in varying degrees before they become numerous. In the Punjab there are two fly seasons, one from March to April, the other from July to October. It is the latter that is generally called the surra season, since equine outbreaks are then generally met with.

The biting flies are not evenly distributed. *Tabanids* are always the most numerous in the following places:—

1. The banks of rivers or nullahs which overflow at certain seasons.

2. Dense jungles in the hills where the rains are heavy.

Tabanus has a very wide distribution, but *Stomoxys* has a still wider. It requires less moisture than *Tabanus*, and where it occurs it is generally in larger numbers than *Tabanus*.

Lyperosia is found in arid camel country, and is dependent on the rainfall to the extent that it swarms in years when there is a good rainfall, while in poor years it is scanty.

Hippobosca camelina is confined to the Trans-Indus portion of the Punjab. *H. maculata* occasionally bites, but does not live on the camel.

The author thinks that in places where bovines are the only reservoir of infection, or where camels seldom come into contact with horses, chance plays an important part in the occurrence of equine outbreaks. The reason for this is that surra oxen generally carry only a few trypanosomes, and only a small proportion of the flies which bite them are likely to become dangerous as disease spreaders. Camels on the other hand frequently have large numbers of the parasite in their blood, but they do not come into close contact with horses.

Once the first horse has become infected, however, the disease spreads rapidly provided circumstances are favourable, because the first horse has large numbers of trypanosomes in its blood and the animals stand together in such a way as to make mechanical

transmission easy. Clinical observations support this view. Surra does not appear in horses in every fly zone annually; if the first case in the horse occurs early the disease generally runs through the stud, while if it be late the outbreak is limited, there being few flies left to effect the transmission.

The circumstances under which camels live make it easier for mechanical transmission to be effected.

The author suggests the possibility that the *T. evansi* may be in reality only a variety of the *T. brucei* which has acquired its characteristics owing to innumerable passages through camels.

- (8) MESNIL (F.) & LEGER (M.). Documents relatifs au Surra des Caprins et à leur Immunité. [Surra in the Goat, and Immunity].—*Bull. Soc. Path. Exot.* 1912. Jan. Vol. 5. No. 1, pp. 31-35.

Although there is ample evidence that ruminants frequently recover from trypanosome infections and thereby acquire a strong immunity, there are but scanty references in literature to the duration of such immunity.

LAVERAN and MESNIL record the case of a goat which having been the subject of nagana for a period of five months was still immune to that disease 21 months later, and was infected in the interval with mal de caderas and surra.

A second goat, as recorded by the same authors, was infected with nagana for a period of four months (Dec.-April) and with surra from Oct.-March. More than a year after its recovery from nagana its serum proved protective for a mouse against the *T. brucei*. The serum of this goat was again tested and found to be protective after 18 months.

According to WRYBURG two zebus which had recovered from surra in Sumatra were immune for a period which did not appear to exceed two years.

MESNIL records that two bovines inoculated at Alfort recovered. Their immunity was tested on three occasions by inoculations with *T. evansi*, to which they proved resistant. One of these animals was again tested a month later with the Mauritius strain and again resisted infection. This not only established the identity of the trypanosomes, but showed that the animal had retained its immunity for a period of more than two years.

LAVERAN records the persistence of immunity in a goat, which had been infected with *T. evansi*, for 2 years and 4 months, and in a sheep which had recovered from a dimorphon infection for 22 months. The serum of this sheep taken 8 months after recovery protected mice in doses of 0.25 cc. He also mentions a goat which recovered from a dimorphon infection and whose serum proved protective 17 months later.

Finally the immunity of a *Macacus* against the *T. gambiense* may persist for more than a year.

The authors record the following experiments of their own.

Goat S.—Inoculated Nov. 26th 1907 with *T. congolense*; infection lasted about 5 months. Reinoculations practised 8 and 12 months later proved negative.

In Jan. 1909 the animal was inoculated with Indian surra, with the result that it was infected for a period of about 4 months. Mice and a dog inoculated from it before May 1909 were infected, but a dog inoculated in June remained uninfected.

Goat J.—Inoculated with Indian surra at the end of January 1909 became infected and recovered in about 4 months. The results of the inoculation of test animals were the same as in the preceding case.

Goat R.—Inoculated with Taher virus of Algerian horses (Ed. and Et. SERGENT). The animal contracted an infection which lasted for 3 months. Nine months later it was infected with the same virus and again recovered.

Goat B.—Inoculated in May 1909 with a strain derived from Senegal. The infection lasted for more than six months, and a number of animals (mice and a dog) inoculated from it before December died from the disease; mice and a dog inoculated in March 1910 did not react.

Before the goats were reinoculated with Indian surra in May 1911, the protective power of their serum was tested. In the case of two of the animals the serum was found to stay the course of the disease slightly when injected into the peritoneum of mice together with the *T. evansi*. The serum of the other two was without any protective effect.

The inoculation of the goats shewed the absolute concordance between the protective power of their serum and the immunity acquired by them.

Animals S and J, which had recovered from surra about 2 years previously, and the serum of which retarded the disease in the mouse, did not become reinfected, nor did animals inoculated from them. The other two (R and B) became infected. Trypanosomes were found in the blood of goat R, but not in that of goat B.

The duration of the disease in these two goats was determined by the inoculation of experimental animals. In the case of goat R it was found that dogs and mice inoculated in November of the same year did not become infected.

Goat B.—One mouse inoculated on the eighth day after the goat was inoculated remained healthy, but a number of mice and a dog inoculated subsequently contracted the disease. The period of incubation was long and the disease developed slowly; parasites were not constantly present in the peripheral blood. Animals inoculated in November failed to react.

Two of the goats originally inoculated with surra did not react, the other two giving positive results. With regard to goat R, it is known that the trypanosome of Taher is different from that of surra. The result in the case of B was more unexpected, since the trypanosome used for the primary infection has been shewn by one of the authors to belong to the species *evansi*. In any case the result would not invalidate the result obtained in the first case. The goat had not any greater immunity against the Mauritius strain. A difference in the condition of the two animals immediately before inoculation may have had some

influence on the marked difference in the infection in the two cases. The authors point out, however, that the disease in the two goats lasted a similar length of time, and suggest that something may be learnt from it regarding the difficult problem of the identification of viruses.

Three months after the inoculation with Indian surra the serum of the four goats was tested as to its protective powers. As in the preceding cases this was done by inoculating mice intraperitoneally with $\frac{1}{2}$ cc. of serum mixed with $\frac{1}{10}$ cc. of citrated mouse blood containing the *T. evansi* (Indian and Mauritius strains).

All the mice so treated were living at the end of 50 days, the controls being dead in 4-5.

The protective power of the serum of goats S and J was evidently increased by the reinoculation although the animals did not have any reinfection.

If it be true, at any rate for the goat, that the protective power of the serum is an index of immunity, and the authors' experiments would appear to corroborate this view, it may be supposed that as the serum of goats S and J became more strongly protective as a result of the reinoculation, the immunity of the animals also increased. Up to the time of writing the protective power of the serum of these two animals had remained at the same level.

Goats S and J were reinoculated in September 1911, the first with nagana (Uganda strain) and the second with the Zululand strain. In both animals there was a slight, but at the same time distinct reaction. The authors finally state that the serum of these goats proved protective against the homologous nagana, but not against the heterologous.

- (9) LEESE (A. S.). Third Series of Experiments on Treatment of Surra in Camels, with some Cures.—*Jl. Trop. Veter. Science*. 1912. Jan. Vol. 7. No. 1. pp. 1-18.

The author refers to the report of the second series of experiments, which was published in the *Journal of Tropical Veterinary Science*, 1910, Vol. 5, p. 397, and which shewed that neither white arsenic nor yellow orpiment given in bolus had any trypanosome-dispersing effect; but that atoxyl (intravenously), tartar emetic (intravenously) and sodium arsenate (in solution per os) are all useful. The dosage necessary for dispersing trypanosomes with these drugs was ascertained and combined treatments shewed fairly promising results, the return of the trypanosomes into the circulation being delayed as long as from 44-58 days after the last dose.

The present series of experiments was carried out principally with these three drugs.

The atoxyl solutions were always freshly prepared and the sodium arsenate was the amorphous salt.

For convenience of description the author indicates the different methods of treatment by applying to them the number of the first camel cured in each case.

“264” *Method*.—Four grammes to 4.4 of atoxyl intravenously, 0.5 gramme of tartar emetic, and 55 to 60 grains of sodium arsenate in drench were given on three consecutive days at intervals of 2, 4, 8, and 12 days.

Three camels shewed no relapses in from 253-498 days, and one relapsed 19 days after the last dose.

The blood was examined daily, and had been examined for a period daily before treatment was commenced, in order to exclude from the treatment any animal in the process of natural recovery.

Although the author does not attach any great importance to the inoculation of small animals with blood of camels not shewing trypanosomes, when the results of such inoculations are negative, nevertheless a guinea-pig was inoculated from each of the recovered animals with negative result.

One of the camels was inoculated again with surra on the 498th day after the cessation of treatment and again took the disease, shewing that the animal was cured of the first infection by drugs; for had the recovery been a natural one there would have been immunity.

Remarks on the treatment.—The treatment is a reducing one, and almost amounts to keeping the animal in a state of chronic poisoning for a time.

The objection to the treatment is its length, and it cannot be considered as suitable for camels in poor condition.

No fatal case of poisoning occurred, but in one where the treatment was a little different in that 4.4 g. of atoxyl were given in the first two series of doses instead of 4 g. poisoning resulted.

In a second case the first dose of 4 g. of atoxyl failed to disperse the trypanosomes in 24 hours, so the dose was repeated on the following day before the antimony was given. Atoxyl poisoning resulted. It appears to be impracticable to omit the last series of doses with the idea of shortening the treatment.

“643” *Method*.—In this experiment the same drugs were used as in the foregoing, but they were given in rotation at intervals of one day, the last dose of sodium arsenate being given on the 27th day. Only one camel recovered, and it is possible that certain dosage experiments carried out previous to the treatment of this animal may have assisted in its cure.

No case of atoxyl poisoning occurred. The blood of the recovered animal was examined for 535 days without any trypanosomes being found and experimental inoculation was also negative.

With the idea of modifying the “264” method, experiments were made with regard to the dosage of atoxyl administered subcutaneously. It was found that 9 grammes caused slight anorexia for a few days, and that 12-18 g. caused poisoning.

Atoxyl poisoning appears to occur more readily if the drug be given with sodium arsenate than when given alone. It is also caused by the repeated administration of doses that would not cause poisoning if given singly.

“588” *Method*.—The treatment was the same as the “264” method save that 5-6 g. of atoxyl were given subcutaneously. The three drugs were administered three times and intervals of 3 and 7 days were allowed to elapse between each series of administrations.

Five animals were treated. One recovered which had relapsed under the “643” method. One died from disease not referable to the treatment, and three relapsed in 12-69 days.

“436” *Method*.—This resembles the “588” method, but four series of doses were administered and the dose of atoxyl used was 5-7.5 g.

Time has not yet elapsed to allow any statement of the full results, but the method appears to be open to grave objection owing to the likelihood of poisoning.

“957” *Method*.—In this method the sodium arsenate was administered before the atoxyl, with a view to getting the effects of the two arsenic compounds about the same time. The treatment however appears to be too dangerous.

Experiments showed that the dose of tartar emetic could be increased with safety. The author used up to 3 grammes, but found that 1.5 g. was about the limit of safety.

“178” *Method*.—This method closely resembles the previous one and is as follows:—

1st day—55 grains sodium arsenate per os.

2nd day—5 grammes atoxyl subcutaneously.

3rd day—1.5 g. antimony tartrate intravenously, followed by a repetition of the series after an interval of three days.

It is a good thing to give 2 lbs. of Mag. sulph. 12 hours after the last dose as some camels incline towards atoxyl poisoning.

The first animal treated had no relapse and ten others under the treatment are not yet ready for report.

“203” *Method*.—This is exactly the same as the “643” method save that the dose of antimony is altered to 1.5 grammes. One animal has been treated and has not relapsed (6 months), and there has been a great improvement in condition.

The irritant Effects of Sodium Arsenate.—Of the total number of experiments in which sodium arsenate was given in full doses combined with other drugs 12 per cent. have died owing to its irritant effects, apart from cases of nephritis.

Full doses of sodium arsenate have been given alone in dosage experiments 25 times without fatality. A “full” dose is one that is fairly reliable for the dispersal of trypanosomes.

Experiments are being carried out with smaller doses.

HOLMES’ “arsenic alone” treatment was found to be impracticable in the camel. White arsenic alone had no effect on the trypanosomes; if given with an equal weight of potassium carbonate it disperses the trypanosomes, but is exceedingly irritant.

- (10) BREISINGER (K. A.). *Chemotherapeutische Versuche bei experimenteller Trypanosomiasis der Rinder*. [Chemotherapeutic Experiments on Cattle experimentally infected with Trypanosomes.]—*Zeit. f. Hyg. und Infektionskrankh.* 1912. April 25. Vol. 71. No. 3. pp. 367-403.

The experiments were undertaken to ascertain (1) whether certain chemical substances were capable of causing immediate and permanent disappearance of trypanosomes by a single intravenous injection; (2) how the animals withstood doses which achieved this object.

All the animals used were inoculated with nagana, the strain being one that was slightly virulent for cattle, but which killed mice in 3-5 days.

Ox 1 was treated with arsenophenylglycin and tartar emetic. Oxen 2 and 4 were treated with salvarsan, and No. 10 with a new dye named "Trypaflavin B," which together with the salvarsan was obtained from EHRLICH.

All the animals were examined for flagellates both by culture and by experimental inoculation before being used for the experiments, with negative results.

A somewhat extensive review of the literature is given.

Ox 1.—This animal was infected by inoculation with the heart blood of a mouse infected with nagana. Trypanosomes were demonstrable in the peripheral blood on the 5th day after infection.

Treatment was begun on the 8th day, trypanosomes being then visible in the blood.

A dose of 18.55 grammes of arsenophenylglycin was injected intravenously in 600 g. of sterile salt solution, the solution being heated to 38° C. before injection. Three-quarters of an hour later 5.3 g. of tartar emetic were administered in the same way dissolved in 530 g. of distilled water heated to 37° C.

The animal very shortly showed signs of uneasiness and colic, and died 2 hours later.

The arsenophenylglycin was administered in the proportion of 0.035 g. per kilog. body-weight, and the emetic in the proportion of 0.01 g. The principal lesions found were: Oedema of the lungs and numerous blood clots in the small bronchi. In addition to these there were numerous haemorrhages in the various tissues and organs.

From the descriptions given in literature by other authors it would appear that death was caused by the arsenophenylglycin, it being questionable whether the tartar emetic was in any way responsible.

Mice inoculated at the time when the post-mortem was made—four hours after death—remained healthy and free of trypanosomes, and since motile trypanosomes are generally discoverable in the blood for a much longer time after death it would appear that the treatment had the effect of clearing the blood stream of parasites.

Treatment with Salvarsan (Ehrlich). Ox 2.—This animal was inoculated with nagana as in the case of No. 1. As no trypanosomes were visible in the blood by the 10th day a mouse was

inoculated with blood drawn from the ear and the animal was also reinoculated. The inoculation of this mouse showed that as a matter of fact the first inoculation was successful. Trypanosomes were found to be present in small numbers on the 5th day after the second inoculation. Mice were also inoculated after the second inoculation of the ox. The mice inoculated after the first inoculation died on the 12th and 16th days, showing that the trypanosomes were very scanty in the blood. Those done after the second inoculation died in 5-6 days.

On the 32nd day after the first infection parasites were found to be numerous in the blood and treatment was resorted to.

Salvarsan was administered in the proportion in which it is administered to the human subject, *i.e.*, 0.01 g. per kilog. body-weight. The solution used was prepared in the following way:—

4.4 g. of salvarsan were taken from vacuum tubes, in which it is kept protected from the light, and mixed with 400 cc. of 0.9 per cent. sterile salt solution, the mixture being agitated until solution was complete. To this were added 8.27 cc. of standard caustic soda solution. The resulting precipitate was dissolved by the addition of a few drops of caustic soda save for a few flakes which settled to the bottom. To the clear solution 600 cc. of sterile salt solution were then added. The mixture was warmed to 37° C. and then injected slowly into the left jugular vein. The actual quantity of salvarsan introduced was only 4.3 g. since there was a little precipitate.

Three-quarters of an hour after the injection there were severe muscular rigors of the body and limbs, the animal had an anxious expression and there was straining, and the animal went down.

An hour and a half later the animal had become quite quiet again and the rigors had absolutely disappeared.

The temperature rose to 40.2° an hour and a half after the injection and then gradually sank to normal again. The animal appeared to be quite lively 7½ hours after injection and its appetite was good. There was no evidence of local inflammation.

Test of the therapeutic Value of Salvarsan Treatment—

Mice were inoculated 3 and 6 hours after the injection of the drug, films and moist preparations being also examined.

Between the 23rd and the 72nd hours after the injection 9 thick preparations from which the corpuscles were removed by solution, 13 blood films, and 8 hanging-drop preparations were examined.

From the 4th to the 10th day the blood was examined daily, and then fresh blood and thick preparations were examined every 3rd or 4th day. Eight days after the injection of salvarsan broth tubes were inoculated with defibrinated blood and two mice were inoculated.

None of the mice showed trypanosomes in its blood, and all remained healthy for four months.

No trypanosomes could be discovered in the films or moist preparation made 3 and 6 hours after treatment. There were immense numbers of collections of round blue-stained granules in practically every preparation, which the author considers to be remains of the disintegrated bodies of the trypanosomes.

Further examination of blood smears and the inoculation of control mice showed that trypanosomes did not reappear in the blood of the ox during the whole period that it was under observation (16 weeks). Five and a half weeks after the treatment the author discovered in the blood rounded bodies with or without a flagellum, and some with two flagella which he believes to be connected in some way with trypanosomes.

Ox 4.—After the blood of this animal had been examined for trypanosomes or “culture flagellates” with negative results, it was inoculated intravenously with 30 cc. of culture medium containing “culture flagellates” obtained from a calf.

This inoculation was a failure.

The animal was then inoculated with the heart blood of two mice infected with trypanosomiasis.

Trypanosomes were discovered in the blood on the 3rd day. On the 6th day broth cultures were made and on the following day no trypanosomes could be found in the tubes. There was no subsequent development of “culture flagellates” in the tubes.

As the number of trypanosomes in the blood fell off, the ox was reinoculated intraperitoneally with the heart blood of three trypanosome-infected mice.

Trypanosomes were numerous present in the blood on the 12th day after the first infection.

This animal was treated in the same way as No. 2 save that the dose given was half that per kilogram body weight.

There was a rise of temperature and some general disturbance.

Mice were inoculated and blood preparations examined as in the case of Ox 2.

In thick preparations made 2, 3 and 6 hours after treatment there were observed large numbers of well defined rounded trypanosome forms, some of which had no flagellum and others had one or two.

By the 19th hour these bodies were scantily present, but they were found 28 hours after and also on the 2nd and 3rd days. No trypanosomes could be discovered.

No trypanosomes were found in the inoculated mice, nor were rounded forms observed although they were present in the blood used for inoculation.

Numerous trypanosomes were found in the blood of a mouse inoculated from the ox 9 weeks after treatment, and dead on the 9th day. There was therefore a relapse.

Treatment with Trypaflavin B (Ehrlich). Trypaflavin is a light crystalline cinnabar-red powder that is fairly soluble in salt solution.

In hanging-drop preparations containing one drop each of 0.4 per cent. trypaflavin solution in 0.9 per cent. salt solution, prepared an hour previously, and of mouse blood containing large numbers of nagana trypanosomes, the parasite exhibited at the end of ten minutes only sluggish movements and rosette clumps were formed. After 15-20 the trypanosomes were practically motionless and after 30 minutes quite motionless.

Ox 10.—Before infection neither trypanosomes nor culture flagellates could be discovered in the blood.

The animal was infected with the heart blood of a mouse in which trypanosomes were numerous.

From the 4th-7th day after infection rounded flagellate and non-flagellate forms were discovered in thick preparations. From this day on up to the time of treatment trypanosomes were present in the blood. No culture flagellates were found in cultures made on the 8th day after infection, nor were nagana trypanosomes discoverable in the tubes two days later. In order to test whether the milk contained trypanosomes two mice were inoculated with 1.5 cc. of centrifuge deposit but both remained healthy.

The dose of trypanflavin used was 0.01 g. per kilog. body-weight.

A 0.4 per cent. solution was made by simply shaking the powder up with salt solution warmed to 40° C.

The solution was injected into the jugular veins, but owing to the restlessness of the animal a portion escaped into the subcutaneous tissue and a portion was lost. The animal probably received only about 3.5 g. of the trypanflavin.

Immediately after the injection an egg-like swelling about 10 centimetres long formed at the seat of operation.

There was no rise of temperature, but from time to time there were rigors.

On the following day there was on the right side a hard painful cord, principally in the connective tissue but also involving the jugular vein, about 20 centimetres broad and 8-10 thick. At the seat of injection on the left side there was a swelling about the size of a man's fist. The swelling extended until on the 10th day after injection the inferior border of the neck was quite rounded and the trachea surrounded. At its lower end the swelling was sack-like. The head was held low and extended owing to the weight and immobility of the neck.

The animal was handed over to the clinique for treatment, and the application of a cantharides ointment caused a decrease in the size of the swelling by the 4th day. Subsequently there was only a little thickening of the tissues remaining.

Mice inoculated 3 and 9 days after treatment remained healthy. Of the two inoculated 6 hours after treatment one remained healthy and the other became infected and died on the 11th day.

In smear preparations made 2 hours after treatment there were numerous collections of blue granules, which sometimes showed red-stained granules among them, and also the remains of trypanosomes in the form of collections of blue-stained granules arranged in a row and containing an obvious nucleus and centrosome. No intact trypanosomes were found.

In a thick preparation made 31 hours after treatment there were found three solid-looking rounded forms in a clump, having a darkly stained protoplasmic body, a large and a small red-stained nucleus, and a single flagellum. On the second day after treatment a vacuolated body of an oval shape with a large red-stained nucleus was found. Non-flagellated forms were found 20 days after treatment, and uni- and biflagellate forms were found after 4 weeks.

Trypanosomes were subsequently proved to be present in the blood by experimental inoculation of mice.

Culture flagellates were proved to be present in the blood of this animal in the summer of 1910 and also 1911, but they disappeared during the winter.

Infection Experiments with Oxen 3 and 6.—The experiments carried out with these two animals deserve special mention on account of the peculiar results obtained.

Ox 3 had been previously infected successfully with nagana three times and Ox 6 once. The animals recovered spontaneously.

Ox 3 was inoculated with nagana from a mouse, but the inoculation failed as shown by inoculation of a mouse. The inoculation was repeated a fortnight later with similar results, but 9 days after the second inoculation the author discovered in thick preparations oval bodies which in many instances were provided with two flagella and which possibly stood in some relationship to trypanosomes.

Thinking that the negative results of the nagana inoculations were due to the low virulence of the strain used, the author again inoculated the animal with blood both subcutaneously and intravenously from Ox 10 which had large numbers of parasites in its blood at the moment.

Six days later round flagellated bodies were found in thick preparations, but neither culture flagellates nor trypanosomes could be demonstrated in the blood either by inoculation or by culture. The animal was finally inoculated intraperitoneally on two occasions with 50 cc. of blood from Ox 4, there being large numbers of trypanosomes in the blood at the time.

Again the animal did not react, but from the 2nd to the 5th and again on from the 10th day flagellated and non-flagellated bodies were found in thick preparations.

Culture trypanosomes had been found in the blood of Ox 6 during the summer of 1910 and this animal was used to test its susceptibility to the trypanosomes found in the blood of German cattle by BEHN. It was accordingly inoculated intravenously with 75 cc. of defibrinated filtered blood diluted with salt solution, the blood being taken from Calf 11.

The blood was examined for four weeks, but neither culture trypanosomes nor blood trypanosomes could be demonstrated by any method.

Repeated attempts, similar to those made in the case of Ox 3, to infect Ox 6 with nagana failed, but on the 3rd day after the first attempt and from the 1st-6th days, and again on the 10th day, numerous flagellated round forms were found in thick preparations.

Experiments to infect mice and hedgehogs with the rounded bodies failed, but it is worthy of notice that subsequently culture flagellates were again demonstrable in the blood of Ox 6 by cultivation.

The author believes that the failures to infect these two animals were due to immunity acquired by the previous inoculations.

Experiments showed that the serum of these animals conferred no appreciable immunity on white mice.

The author gives a brief survey of the literature regarding the rounded forms and then gives details of those observed by him. They varied from 6-10 microns in length and 4-6 in breadth. The cytoplasm showed numerous vacuoles and stained pale blue. The nucleus varied greatly in size, the largest being about 4 microns. The protoplasm contained in many instances numerous small chromatin granules, which were frequently surrounded by a clear zone. The blepharoplast was recognisable in many cases only by its dark carmine tint.

In the forms showing two flagella both of these came off from one side of the body, and very frequently one of them occupied a position around the edge of the body. This was also observed in the uniflagellate bodies. Specimens were observed with four flagella. The bodies were observed in the blood both in cases in which treatment had been resorted to and in cases in which no treatment had been applied.

The author is of the opinion that the rounded forms are fully-grown or dividing trypanosomes which owing to unsuitable conditions have undergone regressive morphological and functional changes.

It is worthy of note that the rounded forms were never observed by the author in thin blood smears, but only in thick preparations.

Conclusions.

I. Chemo-therapeutic treatment of experimental trypanosomiasis in cattle infected with the "Ferox" strain of nagana.—

1. Arsenophenylglycin in combination with tartar emetic is dangerous owing to the large dose required to ensure a cure. The lesions found in animals dying as a result of the treatment are oedema of the lungs, blood clots in the small bronchi, and haemorrhages in the organs and tissues.

2. Salvarsan caused the immediate disappearance of the parasites from the blood stream in Oxen 2 and 4. There was no evidence of intoxication or local reaction in these animals. In Ox 2 there was a very rapid rise of temperature which sank within 2 days to normal.

Nagana trypanosomes were not found subsequently in the blood of Ox 2, which received salvarsan in a dose of 0.01 g. per kilogram body-weight. There was a relapse in the case of Ox 4, which received only half this dose of the drug.

3. Trypaflavin B is of little value owing to the severe local inflammation caused by the injection.

II. Inoculation of Ox 4 with culture flagellates failed entirely, neither blood trypanosomes nor culture flagellates making their appearance.

III. Inoculation of Ox 6 with blood of a calf in which neither blood trypanosomes nor culture flagellates could be demonstrated, but which had been proved infective by inoculation, failed.

IV. Oxen 3 and 6, which had spontaneously recovered from infection with nagana (Ferox strain), failed to react to the same strain, when inoculated with mouse or ox blood, and their immunity was thereby increased.

V. The rounded bodies found in thick preparations of the blood of all the animals, treated and untreated, should be considered as either adult forms or dividing trypanosomes which have undergone regressive changes of morphology and function owing to unsuitable conditions. Trypanosomes could not be demonstrated either in mice or hedgehogs inoculated with ox blood containing these parasites or in broth cultures.

- (11) WALKER (E. L.). The Schizogony of *Trypanosoma evansi* in the Spleen of the Vertebrate Host.—*Philippine Jl. of Science*. Sec. B. [*Philippine Jl. of Trop. Med.*] 1912. Feb. Vol. 7. No. 1. pp. 53-62.

Mention is first made of the descriptions given by SALVIN-MOORE and BREINL (1907) of developmental stages of the *T. gambiense* in the lungs of infected rats. According to these authors peculiar bodies are formed at or near the maximum multiplication of the parasites in the circulating blood. They describe in the process of their formation the development of a stainable band connecting the nucleus with the centrosome, which subsequently disappears. This is followed by the extrusion of the nucleus, the rest of the trypanosome disintegrating. The oval bodies formed by the nucleus and a small amount of protoplasm are stored in the spleen and bone marrow and are termed "latent bodies." These latent bodies are believed by the authors to be specially resistant forms of the parasite which give rise to new generations. The work of CHAGAS (1909) is next referred to. This author published an account of a new trypanosome of man in Brazil which is transmitted by a biting bug (*Conorhinus megistus*). This parasite is named the *Schizotrypanum cruzi*.

It is said by the author that this trypanosome does not multiply by simple division in the peripheral blood, but undergoes a process of schizogony in the capillaries of the lung.

This reproductive process takes place at the times when the parasite becomes numerous in the blood. The trypanosome sheds its undulating membrane and flagellum, bends upon itself, and becomes fused into a rounded or oval body. In some of the parasites the centrosome is shed with the flagellum, and in others it is retained in the body. By division of the nucleus of the first form and of the nucleus and centrosome of the second, and by differentiation of the protoplasm, there are developed from these bodies schizocysts containing 8 small club-shaped merozoites. These escape and, penetrating red blood corpuscles, develop into trypanosomes which make their escape from the corpuscles when mature. The merozoites which have no centrosome develop into female parasites, having a small centrosome derived from the nucleus, and a broad body containing a round nucleus. Those possessing a centrosome develop into male trypanosomes having a large centrosome, an elongated nucleus, and a slender body.

In 1910 HARTMANN found in a section of lung of a guinea-pig infected with this parasite greatly hypertrophied endothelial cells containing large numbers of more or less pyriform bodies with two nuclei. According to Hartmann, similar intracellular stages were subsequently found by Chagas in the heart muscle and brain of a man dead of the disease. He believes that the intracellular stage represents real schizogony, while the extracellular stage with sexual differentiation is to be considered a gametogony.

CARINI (1911) has described a similar schizogony of *T. leptodactyli* in endothelial cells from the heart blood of an infected animal.

FANTHAM (1911), working with *T. gambiense* and *T. rhodesiense*, has confirmed the observations of Salvin-Moore and Breinl regarding the presence of the rounded bodies in the lungs, but disagrees as to their method of development. According to Fantham, from observations made on circulating blood under the microscope, the anterior end of the trypanosome disintegrates, the centrosome migrates to near the nucleus, and the posterior end of the trypanosome with the remnant of the flagellum is cast off, the remainder forming the latent body. He was also able to observe the change of the latent bodies into trypanosomes by mixing infected spleen pulp with fresh blood of an uninfected rat, and observing it under the microscope. He claims also to have transmitted the disease by the inoculation of spleen pulp containing latent bodies, but no motile trypanosomes.

LAVERAN (1911) concludes that the rounded bodies are involution forms.

BUCHANAN (1911) has observed similar bodies to those already described in the organs of the gerbil infected with the *T. brucei* (*pecaudi*), but his explanation of their development is different. The trypanosome bends upon itself and becomes fused, the flagellum remaining attached for a time. Buchanan found in smears from the spleen all stages from the ring-formed parasites to the fairly mature trypanosomes in the red corpuscles.

In the development of the *T. evansi* in the guinea-pig there occur round non-flagellated forms in the spleen and marrow which correspond to the latent bodies described by the other authors, but they are developmental rather than latent, in that they undergo a process of schizogony comparable to that occurring in the *Schizotrypanum cruzi*.

The author has studied two strains of the trypanosomes, one from a horse and the other from a carabao. The strains have been carried on in guinea-pigs, and the development described is based on the examination of the blood and organs of that animal.

The blood taken from the ear veins, and the internal organs of guinea-pigs killed at different periods of infection, have been studied fresh, in dried smears, in smears fixed wet, and in paraffin sections. These preparations have been stained with Giemsa's stain, aqueous alum haematoxylin, Mallory's ferric chloride haematoxylin, and Seidelin's iron haematoxylin. Air-dried smears of both the blood and organs, stained for 12-24 hours

with Giemsa's stain have given the best results. Sections have been useful in determining the relation of the stages of the trypanosome to the tissues.

T. evansi, when inoculated subcutaneously into guinea-pigs, has an incubation period of 5-16 days. The disease runs a more or less chronic course of one to several months duration. The course of the disease is marked by alternating phases of increase and decrease of parasites in the peripheral blood.

It has been found that large numbers of rounded binucleate bodies appear in certain organs at or near the maximum increase of the parasites in the blood. Few or no round bodies could be found during the periods when the parasites were scanty in, or absent from the blood.

The situation of development of these round forms of the *T. evansi* does not correspond with that of the latent bodies of *T. gambiense* and *T. rhodesiense*, nor with that of the schizonts of the *Schizotrypanum cruzi*. The latter are described as occurring in the capillaries of the lungs, but in the case of the *T. evansi* the round forms develop principally in the spleen, and to a less extent in the bone marrow. Sections show that they are developed extracellularly in the small capillaries.

The author's view with regard to the formation of the rounded bodies differs from those expressed by the other authors mentioned. The parasite bends round until its ends meet and then fuses into a rounded body, or into a ring which subsequently becomes solid. The flagellum remains attached around one side of the parasite for a time, but disappears very soon.

The bodies measure 2-5 microns in diameter and have a body, which stains pale blue with Giemsa, containing a central or eccentrically placed nucleus and a centrosome which is often on the opposite side of the body to the nucleus. The nucleus and centrosome stain red with Giemsa. The author has not been able to distinguish the type without a centrosome, as described by Chagas.

In the spleen of guinea-pigs killed when the blood is swarming with trypanosomes a further development of these bodies is evident. The round binucleated body increases in size, and both nucleus and centrosome undergo a process of division. The fully developed schizonts measure 10-15 microns in diameter. The number of nuclei and centrosomes varied from 4-16. Some of the large parasites show evidence of fission of the protoplasm, and others complete differentiation of merozoites, which appear to be surrounded by a thin cyst wall.

The merozoites are arranged like the segments of an orange with a slight spiral twist. They are elongated, sickle-shaped bodies, 6-10 microns long and 1-1.5 wide. They have a central nucleus and a centrosome near one end, but no flagellum or undulating membrane. Forms corresponding to the intracorpuseular stages of Chagas and Buchanan are observed, but the author has not been able to convince himself that the parasites are really within the corpuscles.

In the author's view the merozoites develop directly into trypanosomes.

No sexual processes have been observed preceding or during the formation of the round bodies, nor during the development of the multi-nuclear cysts. Sexual reproduction should, according to the accepted theory, take place in the body of the invertebrate host. The merozoites show no dimorphism, nor has the author observed any sexual differentiation in the mature trypanosomes; he therefore designates the reproductive process a schizogony.

The author is of the opinion that the genus *Schizotrypanum* may have to be abandoned. The schizogony described by Hartmann and Carini is probably phagocytosis of young schizonts by endothelial macrophagi. The absence of simple division in the peripheral blood would appear to be distinctive of the *S. cruzi*; but in view of the fact that this parasite appears to multiply by simple division in the gut of the invertebrate host and in cultures on Novy and MacNeal's medium, a suspicion is raised as to the accuracy of the observation.

Possibly some of the young schizonts persist through the latent period and undergo schizogony at the beginning of the relapse.

Conclusions.

"In the developmental cycle of *Trypanosoma evansi* a schizogony takes place in the spleen of the vertebrate host.

"The observations of Salvin-Moore and Breinl, Fantham, and Buchanan that forms similar to the young schizonts of *Trypanosoma evansi* occur in the internal organs of animals infected with *Trypanosoma gambiense*, *T. rhodesiense*, and *T. brucei* make it probable that schizogony is a reproductive process common to the trypanosomata.

"The validity of *Schizotrypanum* Chagas as a genus distinct from *Trypanosoma* Gruby appears to be doubtful.

"Further investigation is necessary to determine the significance of this schizogony in the life-cycle of the trypanosomata and its relation, if any, to latency in trypanosomiasis and to relapses after chemotherapeutic treatment."

- (12) MASON (F. Eugene). Equine Trypanosomiasis in Egypt.—*Jl. Comp. Path. and Therap.* 1912. June. Vol. 25. No. 2. pp. 93-109.

Having heard it said by the Bedouins that horses after being bitten by flies sometimes developed a disease which presented symptoms identical with those observed in camels affected with el debab, and which was invariably fatal within a few months, the author kept a watch for such cases. The first case encountered was in an Arab horse, the property of the Khedivial Agricultural Society, which was admitted into the Cairo Infirmary. Trypanosomes were easily discoverable in the peripheral blood, and there was also agglutination of the red corpuscles, described by GAIGER as occurring in the blood of horses suffering from surra.

The animal, which appeared to be in good health at the time of purchase, began to lose flesh and to show an irregular temperature some fifteen days later. It was stated that repeated blood examinations had been negative, and that it had been treated with quinine for piroplasmosis. At the time of admission to the Infirmary the animal was very emaciated and weak, the respiration was rapid, the conjunctiva yellow and ecchymosed

and there was oedema of the chest and limbs. Trypanosomes were found in the blood on the following day.

Treatment was commenced on the next day. Two doses of atoxyl followed by orpiment and sodium arsenate were given, the drugs being used in rotation on alternate days, leaving one day's interval. The temperature fell to normal with the first dose of atoxyl. Treatment was continued for about three weeks, when symptoms of arsenic poisoning were observed. In view of the fact that on analysis the orpiment that was used was found to be a mixture of the red and yellow varieties, treatment was discontinued till fresh samples of true yellow orpiment could be obtained from Europe. The animal improved for about five weeks and showed no rise of temperature. When the temperature rose trypanosomes could not be detected in the blood, but there was marked agglutination of the red corpuscles.

The treatment was recommenced five weeks later and was continued for a month. Arsacetin, orpiment, and sodium arsenate were used in rotation at intervals of one day. The dose of arsacetin was from 2.5-3.0 grammes, that of orpiment 6.0-7.0 grammes save in the case of the last two administrations when the doses were 9.0 and 10.0 grammes, the latter being given in two doses; the dose of sodium arsenate was from 2.0-4.0 grammes. After this treatment the animal was in excellent condition, and was discharged for duty.

At the time of writing (6 weeks later) the animal was still apparently perfectly well.

It is of interest to note the following points: It is not certain that the animal was infected in Egypt, as it had been recently imported from Syria, and probably came over with a number of camels.

Before treatment was commenced and at a time when the parasites were not numerous, citrated blood from the jugular was used for the inoculation of a number of animals.

Four white rats were inoculated subcutaneously with citrate mixture corresponding to 1 cc. of blood. These rats died with their blood swarming with trypanosomes in from 22-38 days.

On the same day an English gelding was inoculated subcutaneously with 10 cc. of the same mixture (equal parts of blood and citrate).

On the twelfth day there was an elevation of temperature, but the blood was not examined.

The following morning there was a further rise of temperature and there was marked agglutination of the red cells, but no trypanosomes were found.

On the fourteenth day trypanosomes appeared in the blood.

After this the disease advanced rapidly and clinical symptoms appeared, the animal dying on the twenty-seventh day.

At the post-mortem the following lesions were found: general emaciation, gastro-enteritis, enlargement of the spleen which showed numerous haemorrhagic spots, liver enlarged and stiff, lungs oedematous, lymphatic glands oedematous, heart hypertrophied on the left side and dilated on the right, haemorrhages under the endocardium.

An Egyptian mare was inoculated from the above English gelding on the twenty-fifth day of illness when trypanosomes were numerous. Trypanosomes appeared on the eleventh day.

On the forty-first day she was given 8 grammes of the new sample of orpiment to test its toxicity, and a few days later 15 grammes. There was no colic or diarrhoea. Death occurred on the 60th day.

The lesions were much the same as in the preceding case.

A second mare inoculated from the above Egyptian mare died on the thirtieth day, trypanosomes having appeared on the ninth day. Another mare inoculated with the same material died on the thirty-first day, trypanosomes having been observed on the fifth day.

White rats inoculated from the first Egyptian mare died on the fourteenth, nineteenth, and twenty-first days respectively.

Other naturally occurring Cases.—

Case II. Chronic case. Animal had been more or less ill for about five months when trypanosomes were found.

A dose of 3 grammes of salvarsan administered intravenously caused some improvement, but there was a relapse. Two months later 4.2 grammes were administered with great improvement in condition as a result. Temperature had remained normal up to date (2 months).

Case III. Arab horse fifteen years old. Trypanosomes found about a fortnight after admission to the Infirmary. Treatment begun four weeks later. The treatment adopted was the same as in the first natural case—arsacetin, sodium arsenate, and orpiment. Animal greatly improved in condition at the time of writing.

Case IV. No trypanosomes discovered, but marked agglutination of the red corpuscles. After a fortnight's illness the animal was given a little less than 3 grammes of salvarsan. The full dose was prepared, but the injection had to be discontinued owing to the appearance of urgent respiratory symptoms.

There was a marked temperature reaction on the evening of the same day and also on the sixth day after, the latter lasting till the tenth day.

On the twelfth day 100 cc. of 1 per cent, trypanblue were given subcutaneously and the animal shortly afterwards returned to work and has been working and in excellent condition ever since.

Case V. Animal had been ill (fever) on three occasions before admission to the Infirmary. Trypanosomes found a month after admission. Treatment as in Case I.—Recovery.

Case VI. English thoroughbred. Trypanosomes found. Case said to be doing well under treatment.

The fly seasons in Egypt occur in May and June and again in September.

During these months the *Tabanus taeniola* and *Tabanus ditaeniatus* abound in certain known localities. There appears to be no doubt that they are responsible for the transmission of the disease from camels to horses, and also that the infection of

horses takes place occasionally only. No other domestic species is known to harbour the trypanosome.

The Trypanosome.—

In Case I., the origin of which was doubtful, the shortest parasite observed measured $17.38\ \mu$ and the longest $23.8\ \mu$. The width at the widest part was about $1.42\ \mu$. Compared with the measurements of trypanosomes naturally occurring in camels those found in Case I. were both a little shorter and a little more slender. Otherwise there was a very close resemblance. In both cases the body posteriorly to the nucleus was free from granules, and in some specimens the nucleus was prolonged anteriorly by a horn-shaped mass of granules. The undulating membrane was well developed. The cytoplasm extended to the anterior end of the flagellum.

In comparison with the measurements made of trypanosomes from cases of undoubted Egyptian origin those found in Case I. were slightly smaller, but the author suggests that this may have been due to the preparation from which the measurements were made having dried rather more slowly and thus caused more contraction. The fact that in a drawing made under the same magnification with the camera lucida the blood corpuscles appear smaller supports this view.

In preparations made from infected white rats the parasites appeared to be distinctly larger than in those made from the blood with which the rats were inoculated.

It also appeared that passage through a series of horses was responsible for an increase in the length of the parasite, but not in the width.

- (13) WATSON (A.). Dourine—Its Pathogenicity, and a Practical Test of the Efficacy of Drug Treatment, with especial reference to the Action of Atoxyl and Arsenophenylglycin.—*Report of Veter. Director General and Live Stock Commissioner J. G. Rutherford, C.M.G., for Year ending March 31, 1911.* Canada: Dept. of Agriculture. Sessional Paper No. 15c—1912. pp. 151–156.

The paper presents a summary of observations on the pathogenicity of a strain of dourine in horses, and the results of a series of infections with the same strain in which experimental treatment was applied.

Pathogenicity.—The author has endeavoured to follow a plan which he believes to be the proper procedure in the study of any animal trypanosomiasis. He is of the opinion that the disease and its causal trypanosome should be investigated in the following manner. In the first place cases of natural infection should be examined, this being followed by an investigation of the disease set up experimentally in the natural host, and then the experimental infection of unnatural hosts; the entire process being finally reversed. The author is further of the opinion that experimental infection of unnatural hosts is carried to an extreme, and that too high a value is placed upon pure laboratory experimentation.

Throughout his experiments a single strain of the trypanosome was used. The strain was carried on directly from horse to horse, without any intermediary passage through laboratory animals, and was not subjected to any environment other than its natural host.

The strain used was originally isolated from a stallion that had suffered from the chronic form of the disease for about two years, showed all the usual symptoms, and finally died of the disease.

First generation.—Twelve mares naturally infected. Diagnosis was based on clinical symptoms.

Six of the mares were killed when the disease, which was chronic and intermittent in all cases, had been in existence for about a year. Three of the animals died of dourine and three recovered. In the three animals which recovered the disease lasted from twelve to twenty-one months, while the period of survival and recovery was from two to three and a half years. In the fatal cases the disease lasted from two to three and a half years. The trypanosome was first discovered in an animal which survived for three and a half years, the discovery being made in the seventh and eighth months. The trypanosome which was then found was the starting point of the succeeding generations.

Second generation.—A stallion, naturally infected by one of the above-mentioned mares. The animal was under observation for a short time only before its death, but there was a history of the disease having been in existence for about two years.

Four experimental infections.—Two foals, one mare, and one gelding.

In the first foal the course of the disease was fairly rapid, the animal showing marked elevation of temperature, oedema, enlarged glands, paralysis, emaciation; and death took place in the fifth month.

In the second foal and the mare the disease took the following course. First stage—periodicity of trypanosomes in the vaginal fluids and vulvar oedema, local symptoms only. This was followed by a long latent period, which was succeeded by a period of plaque formation, and typical ocular and nervous symptoms. Towards the end of this period trypanosomes were found in each animal in the contents of the local swellings in large agglomerations, and in different stages of phagocytosis. This throws some light on the immunity acquired by these animals.

Recovery was rapid, and at the time of writing had been maintained for two years.

The gelding shewed enlarged glands after three months, and intermittent inco-ordination and paralytic symptoms from the sixth to the twelfth month. Recovery ensued, maintained two and three-quarter years.

Third generation.—Four experimental infections—two fillies and two foals.

In the fillies the disease lasted six to eight months, and there was no evidence of nervous derangement nor disturbance of general health. Recovery.

In the foals the disease was of the severe, intermittent, chronic type. Foal 1—Fourteen paroxysms of fever each lasting about five days, with intermissions of twenty-one days during the first year. Nine paroxysms of six days each at intervals of thirty days during the second year. Remission of symptoms and general improvement of health during the third year; rare brief relapses. Foal 2—Slight elevation of temperature for long periods, and four paroxysms during the first year. Seven, at intervals of about forty-four days and lasting eight days, during the second year. Both foals shewed the usual typical symptoms.

Fourth generation.—One experimental infection of a foal, the course of the disease being similar to the preceding. Recovery indicated in the third year.

Fifth generation.—One experimental infection of a foal. Disease severe throughout. Death in the eighteenth month.

Sixth generation.—One experimental infection of a foal. Disease severe at the commencement after a short period of incubation. Subsequently remissions and intermissions and a period of recovery. Trypanosomes not seen after the third month. Recovery probable.

Seventh generation.—One foal. Disease severe and acute. Death on the forty-first day.

Eighth generation and ninth generation almost exactly reproduced the seventh.

Tenth generation.—Very like preceding, but death delayed till the 101st day.

The above observations are in agreement with the general experience that the disease is more progressive in the stallion than the mare.

The virulence of the trypanosome was increased by the passages.

Throughout the series the most striking feature was the development and intensification of a characteristic type of fever. Until the third generation was reached fever was a rare symptom; in the third and fourth it was associated with alternating paroxysms and intermissions; in the fifth and sixth generations it was still more constant; and in the last generations it was the outstanding feature. The trypanosome was more easily found in the later cases than in the early ones, but it was never found in the general circulation. Save on two occasions, when they were found in clear lymph escaping from punctured lymph vessels in the tail, they were observed only in the fluid contents of oedematous swellings and in preparations from vaginal mucosa.

There was no intensification of virulence for laboratory animals, and it was found to be impossible to carry on the strain satisfactorily in them.

The mortality was about 50 per cent. or rather less.

Treatment.—Horses only were used for experiments in the treatment of the disease, for the reasons that the strain was difficult to keep running in laboratory animals and the drugs employed had been proved to be more or less efficient for the treatment of other trypanosomiasis.

Atoxyl.—1. The drug in comparatively small doses could not be tolerated by a stallion in the very advanced stages of the disease, and was probably contributory to the immediate cause of death.

2. A filly which received three injections during the sixth month of the disease rapidly recovered. Two foals were obtained from this animal without either the offspring or the stallion becoming infected.

3. A gelding shewing marked paralysis received 10 grammes in seven doses. There was temporary improvement only.

4. A young filly in which the course of the disease was marked by regularly recurring paroxysms with intervals of twenty days received two full doses of atoxyl on the tenth and thirteenth day respectively after an intermission. The next paroxysm was not delayed in the least. Parasites, which had been frequently observed up to the time of administration of the drugs, were not again seen for more than three months.

Atoxyl, Mercury Bichloride, and Potassium Iodide.—5. A mare shewing paralysis to a marked degree received two courses of atoxyl, 18 grammes in all, with injections of bichloride of mercury and potassium iodide in the intervals. There was a complete recovery, which at the time of writing had lasted for two and a half years.

Atoxyl, Mercury and Arsenic Iodide, and Arsenophenylglycin.—6. In a young animal a course of atoxyl given during the fourth month of the disease was without effect. The second course of injections, alternating with injections of a solution containing one per cent. arsenic iodide and mercury iodide, during the sixth and seventh months was also without effect; as was also a third course of large doses of atoxyl during the ninth month. A single large dose of arsenophenylglycin was given at the twelfth month and repeated seven weeks later. The disease made no further progress and the relapses became more infrequent.

Arsenophenylglycin, Trypanblue, and Sodium Arsenate.—7. A filly eleven months old and in the fifth month of the disease received 10 grammes of arsenophenylglycin, and, after an interval of thirty-five days, 12 grammes. About a month later there was a febrile period lasting three days. Arsenious acid and iron sulphate administered daily for a month caused a slight improvement. A mixture of arsenophenylglycin 10 grammes, trypanblue 5 grammes, and sodium arsenate 0.5 gramme was then given. There was a period of remission followed by relapses and death.

Arsenophenylglycin.—8. A filly with severe symptoms and persistent fever received 12 grammes of the drug on the eighty-third day, and forty days later a second dose of 15 grammes. The temperature curve was normal after the first dose, but fever returned on the thirty-third day after the second. On the fourth day of the paroxysm 10 grammes were given. Slight elevation after twelve days, followed by intermission of two months. General oedematous urticaria and fever. Without further treatment the animal apparently recovered.

9. A foal developing acute infection received 7 grammes. The symptoms progressed and a second injection of 9 grammes was given ten days later. Death after two days.

Poisoning with Arsenophenyglycin.—10. Experiment to ascertain the largest dose that could be given with safety.

A healthy stallion received a dose of 30 grammes—80 milligrammes per kilogramme body-weight. No immediate effects. Dullness and depression the following day. On the third day spasmodic contraction of the pharyngeal muscles and difficulty in swallowing, associated with thickening of the lips. In the evening of the same day excitement, rigors, nervous symptoms and death.

Dosage and Administration.—The author found that the administration of 70-75 milligrammes per kilogramme was tolerated, but with some risk, and that 60-65 milligrammes were well borne. He also found that the maximum dose that can be given to a lightweight animal may be toxic if given in the same proportion to a large heavy animal. That is to say, as the body weight increases the proportionate dose should be slightly decreased.

In all cases the drug was given intravenously with a syphon, a one per cent. solution being used.

Remarks.—

"The few experiments cited above do not afford evidence, from a practical standpoint, of a satisfactory method of treatment; some cases have recovered under drug influence, others without any treatment being given at all, and there are cases in which the progress of the disease has scarcely abated, or been but slightly modified, by any of the drugs employed except arsenophenyglycin. A single dose of this preparation will cause the rapid disappearance of trypanosomes from the tissues haunted by them for weeks or months previously, oedema and local symptoms will disappear in unison, yet the characteristic type of fever recurs at stated intervals, and though the parasites are never to be seen again the disease may progress to a fatal termination.

"The variations in individual resistance and in different degrees of virulence are no doubt responsible for much of the apparent contradiction in results. It may be mentioned in passing that in some parts of Africa the treatment of sleeping-sickness with arsenophenyglycin has been attended with marked success, while in others it has as signally failed.

"Dourine, while not by any means the most fatal of the animal trypanosomiasis, is one of the most chronic, and apparently the most resistant of any of them to the influence of drug-treatment."

- (14) DUKE (H. L.). The Transmission of *Trypanosoma nanum* Laveran.—*Proc. Roy. Soc.* 1912. April 10. Ser. B. Vol. 85. No. B. 576. pp. 4-9.

The trypanosome was obtained from VAN SOMEREN, who had found it to be the cause of a fatal disease in the neighbourhood of the Sebwe River. It was found to be identical with *T. nanum* both in its morphology and pathogenicity.

Morphology.—Corresponds very closely to *T. pecorum*.

There is no free flagellum visible. Average length about 14 microns.

Pathogenicity.—The original animal received from van Someren—a goat—was in perfect health at the time of writing—7 months

after infection. From this goat 3 monkeys, 3 white rats, 2 puppies, 1 wild pig, 1 sheep, and 1 goat were inoculated. Of these only the sheep and the goat shewed trypanosomes. The sheep was still in good health 154 days after inoculation, the period of incubation having been 10 days. The goat shewed trypanosomes in its blood after 6 days and died on the 64th day as a result of the infection.

Transmission by G. palpalis.—The first six attempts at transmission failed. Whereas in the first experiment the flies were first fed on a goat, in the second they were fed on the sheep mentioned above.

It was found that the flies fed far more readily on the sheep.

Four experiments were made and the flies were placed on sheep, calves, goats, and dogs. One calf which was used in all four of the experiments became infected.

As a result of the dissection of flies used it would appear that the development of the *T. nanum* in the *G. palpalis* commences in the hind gut and extends forward via the thoracic gut and proventriculus until the proboscis is reached. The salivary glands are apparently not invaded by the trypanosome. A fly with a negative proboscis is presumably non-infective, this conclusion being supported by the fact that on three occasions injection of positive proventriculi failed to infect a goat.

It was found that flagellates may be well established in the proboscis by the 25th day after feeding. Results also indicated that the infection of the proventriculus may be merely a temporary invasion, while the flagellates are becoming established in the proboscis.

- (15) FRASER (A. D.) & DUKE (H. L.). An Antelope Trypanosome.—*Reports of the Sleeping Sickness Commission of the Royal Society*. 1912. No. 12. pp. 56-63. With 10 plates. Abstract in *Proc. Roy. Soc.* 1912. April 10. Ser. B. Vol. 85. No. B. 576. pp. 1-2.

A goat inoculated with blood obtained from a bushbuck shot on the shore of Victoria Nyanza showed trypanosomes in its blood ten days later. The trypanosome corresponded morphologically with the *T. uniforme*.

Cattle, goats, sheep, and bushbuck were infected. Monkeys, pigs, dogs, cats, guinea-pigs, and white rats proved refractory. It was shown experimentally that the *G. palpalis* was capable of transmitting the parasite. The flies became infected in 27-37 days, and the infection in the fly was limited to the proboscis. Flies caught in the same district were found to be naturally infected and capable of setting up infection in a goat. *Trypanosoma vivax* appeared in the goat's blood a few days later.

Conclusions.

1. This trypanosome, which is of fairly frequent occurrence among Lake-shore antelope, is *Trypanosoma uniforme*.
2. The available evidence points to *Glossina palpalis* as being the carrier of this trypanosome.
3. *Glossina palpalis* caught on the Lake-shore are naturally infected with *Trypanosoma uniforme*.

- (16) RODHAIN (J.), PONS (C.), van den BRANDEN (F.) & BEQUAERT (J.). Les Trypanoses animales au Bas-Katanga et leurs Rapports avec les Glossines. [Animal Trypanosomes at Bas-Katanga and the Part played by Glossina in their Transmission.]—*Bull. Soc. Path. Exot.* 1912. Jan. Vol. 5. No. 1. pp. 45-50.

Two types of trypanosomes were found: one of the *congolense* and the other of the *cazalboui* type.

1. *Trypanosomes of the Congolense Type.*—Trypanosomes of this type were found in dogs and goats. The disease produced may be acute or chronic. In the dog the acute cases observed terminated fatally within a month, the chronic cases showing the usual symptoms of trypanosomiasis and slight posterior paralysis.

In preparations fixed in osmic acid the average length of the parasite was about 12·5 microns, the width at the level of the nucleus being 1·5.

The trypanosome was pathogenic for the guinea-pig, death taking place in about three weeks.

The majority of cases observed in the goat were chronic. The trypanosome was about the same size. Three guinea-pigs inoculated from two different goats failed to react, as did a young indigenous dog.

Glossina palpalis and *G. morsitans* were both present at the places where two of the diseased dogs and one of the goats were found. The other animals that were found infected had been transferred several months previously to places where the *G. morsitans* was present exclusively. *Tabanus* was present in very scanty numbers, while the tsetses were abundantly present.

The authors were able to obtain evidence that the *G. morsitans* is capable of transmitting trypanosomes of the *congolense* group. Flies were fed on animals affected with trypanosomes of both types and when afterwards examined were found to have immense numbers of trypanosomes in the anterior portion of the mid-gut, and the parasites were swarming in the proboscis.

The authors made no inoculations with material obtained from the infected proboscides, but they hope to furnish laboratory proof that the infection of the flies is a double one. The proportion of flies found to be infected was high—41 per cent.

Treatment Experiments.—Subcutaneous injection of a 5 gm. dose of arsenophenylglycin caused a disappearance of the trypanosomes, but they reappeared in the circulation 15 days later and the goat died in 36 days. Two goats received respectively one and two doses of emetic intravenously, but relapses occurred in both cases. These animals were subsequently treated with trypanosan administered by the mouth.

A complete recovery followed the administration of 8 and 6 g. of the drug, the administration being spread over two days. Two other animals recovered which were treated with emetic followed by trypanosan, but the authors attribute the cures to the dye.

Experiment showed that doses of the dye not exceeding 0·5 g. per kilog. were well tolerated, and it was further found that 0·008 g. of emetic per kilog. could be administered for several

consecutive days without bad results. On the other hand the administration of 0.10 g. of emetic to a dog intravenously was followed by serious disturbance.

2. *Trypanosomes of the Cazalboui Type*.—This type of trypanosome, which is easily recognisable in moist preparations by its rapid translatory movement, was found by the authors in the blood of a number of different species of antelope, an eland, and goats.

The parasites were present in small numbers only save in the case of one animal. Inoculation of goats was positive, but subcutaneous inoculation of guinea-pigs failed. Neither the animals in which the parasites were discovered, nor goats infected from them appeared to suffer as a result of the infection.

The largest trypanosome observed in the blood of an antelope measured 31 microns by 2.1, but the average size was 24.4.

No *G. palpalis* were found near Lake Kabwe, but *G. morsitans* was abundantly present. The observations were made during the dry season. Pangonia had disappeared, and Tabanidae were very rarely found. It is probable that the *G. morsitans* is the transmitting agent. Flies which fed upon a goat infected with the *T. cazalboui*, when afterwards placed on a goat which had recovered from the *T. cazalboui* under treatment with emetic and trypanosan, infected it.

Experiments with the same flies on guinea-pigs failed.

Five of the flies were examined after death and infection of the proboscis only was found in three of them. Subcutaneous inoculation of a goat with a proboscis determined infection after 13 days incubation. Both trypanosan and emetic have very rapid effect on the trypanosome, causing complete disappearance.

The authors note that a recovery determined by drug treatment does not set up immunity.

- (17) LAVERAN (A.). *Expériences d'Immunité croisée avec Trypanosoma brucei, Tr. brucei var. Werbitzkii, et Tr. rhodesiense*. [Cross-Immunity Experiments with *T. brucei*, *T. brucei* var. *werbitzkii*, and *T. rhodesiense*.]—*Bull. Soc. Path. Exot.* 1912. Feb. Vol. 5. No. 2. pp. 101-105.

Two sheep were inoculated with *T. brucei* and *T. brucei* var. *werbitzkii* respectively. After their recovery they were found to be immune to the trypanosome with which they had been infected. They were then inoculated each with the other trypanosome and it was found that the animal primarily inoculated with the *T. brucei* did not become infected with the *T. brucei* var. *werbitzkii*, and that the one inoculated with the latter became infected with the *T. brucei*, but that the infection was a slight one.

It was also found that the serum of the sheep inoculated with nagana (*Werbitzki*) was active when mixed with the acentrosomic *T. brucei* and feebly active with the *T. brucei*. On the other hand the serum of the sheep inoculated with nagana was found to be active in mixture with the acentrosomic trypanosomes and less active with the nagana parasite.

Low's suggestion that the *T. rhodesiense* was simply the *T. brucei* was tested by inoculating both the sheep with *T. rhodesiense*. Both the animals became infected and died.

- (18) DARLING (S. T.). The Pathological Anatomy of Natural and Experimental Murrina—a Trypanosomal Disease of the Isthmus of Panama.—*Jl. of Med. Research*. 1912. June. Vol. 26. No. 2. pp. 219-247.

Murrina, also known locally as Derrengadera and Morina de Cadera, is a disease of equines in the Panama Canal Zone. The cause is the *T. hippicum*. The author gives full details of the post-mortem findings in a number of natural and experimental cases in horses, mules, and ponies, and also the details of the lesions found in a number of monkeys, dogs, racoons, rabbits, guinea-pigs, mice and rats. Further details are also given regarding the histology of the lesions.

The following is an extract of the author's summary.

Murrina is an intoxication resulting in cellular degeneration and necrosis. The continuity of the endothelium is injured or destroyed and as a consequence effusion, oedema, and terminal ecchymoses arise. The toxic agents elicit certain reactions in the host: namely, lymphocytosis, auto-haemagglutination, phagocytosis of erythrocytes and trypanosomes, hyperplasia of the spleen, bone marrow, and lymph nodes, and cellular exudations in the kidney, liver and elsewhere.

The degree of splenic hyperplasia varies inversely with the size of the infected animal, being greatest in the smaller rodents, and is directly proportional to the anaemia and the degree of infection. The hepatic necroses and the collections of lymphoid cells in the liver and kidneys are of special interest. There are two types of necrosis: First, the large, central zone areas, in which the hepatic cells have undergone a hyaline, granular or fatty change, and in which there is usually no leucocytic exudation. Second, the smaller areas found in the intermediate and peripheral zones in which there is a polymorphonuclear and mononuclear leucocytic exudation replacing the parenchyma. Each type of necrosis suggests a different etiological factor. Again, in this infection there is a very marked lymphocytosis and in many of the areas of necrosis the cells may be entirely lymphoid in type or preponderatingly so. This requires further investigation.

The collection of cells in the liver and similar collections in the kidneys are of the polyblast and lymphocyte type, and they can be seen in many locations emerging from the arterioles and capillaries, having come originally from the spleen, in which organ they can be seen on the peripheries of the Malpighian bodies. These cells probably come from the lymph nodes and bone marrow as well. It would appear that the toxic agents in this infection specifically attract and stimulate the production of cells of the mononuclear type.

The cellular collections in the liver necroses are phagocytic in character and represent efforts to remove normal or degenerate hepatic cells.

The blood destruction and stimulation of the blood-forming organs leads to hyperplasia of the yellow bone marrow and to an interesting picture in the spleen of the coati, guinea-pig, monkey, rat and mouse. The spleens of these animals were found normally to contain a few megakaryocytes and nucleated red cells. Eosino-

philes were absent. Yet, when these animals were infected with *T. hippicum* the spleen, particularly that of the coati, took on to an intense degree the picture of red bone marrow, in that the megakaryocytes were greatly increased in number, and eosinophile myelocytes and polymorphonuclear leucocytes, as well as nucleated red cells, were seen. In this animal it would appear that the latent myelopoietic function of the spleen had been greatly augmented. The interstitial collections of cells in the kidneys of the racoon contained here and there some clumps of eosinophilic polymorphonuclear leucocytes, but no megakaryocytes.

In other mammals it was noted that when megakaryocytes normally were not seen in the spleen, the trypanosome infection did not provoke their appearance in any other location than in the marrow of the long bones.

In the experimentally produced disease in mules the autoagglutination of erythrocytes frequently appeared before the trypanosomes could be demonstrated in the peripheral blood.

The lesions in this trypanosomal disease conform with those of other trypanosome infections.

The lesions encountered in horses and mules were constant, though such features as oedema, emaciation, and ecchymoses varied with the stage of the disease. The gross lesions characteristic of the disease in equines are petechiae of the pleura, pericardium, nasal and conjunctival mucosae, peritoneum, renal cortex, and capsule of the spleen. There are haemorrhages of larger size in the endocardium, epicardium, and renal lymph nodes. Emaciation and anaemia are usually constant, though in the fulminating type of the disease the former may not be present. Effusions or localised areas of oedema of the sheath or hock, and particularly a longitudinal strip of the lower belly wall are seen. There is slight splenic enlargement and myeloid hyperplasia of the yellow marrow.

In smaller animals haemorrhages are not nearly so common; in fact, they are generally absent. Splenic enlargement becomes more noticeable and constant in the smaller animals. There was acute iridocyclitis in the dog, racoon, and monkey. Rabbits always presented dermatitis of the ears, and muzzle, and a marginal blepharitis. Ulceration of the scrotum was occasionally seen in guinea-pigs.

Histologically the lesions are distinctive. Nephritis was noted in horses and mules, but was different in type from that seen in the racoons and monkeys. The cellular changes in the liver of the guinea-pig are constant for that animal.

The features of the lesions in horses and mules are the agglutination of the red blood cells and their phagocytosis by endothelium and large mononuclear phagocytes in the liver, lymph nodes, and splenic sinuses; and hepatic necroses of two types, hyaline and inflammatory. Acute glomerulitis and acute haemorrhagic nephritis are very constant, as are the various petechiae and ecchymoses in which there may be polymorphonuclear or mononuclear leucocytic exudation. In the spleen the chief change noted is the very large amount of haemosiderin brought thither by large mononuclear phagocytes.

These changes occur with great rapidity in the small rodents and in these animals a great many trypanosomes are produced in proportion to the size of the animal in so short a time that they are rapidly overwhelmed. Trypanosomes are never as numerous in equines as in the small animals. Though equines are ultimately killed by the infection the lesions presented are, excepting the associated anaemia, very acute.

The histological changes in the smaller animals are generally more extensive, when similar in type, than those occurring in equines. The spleens show hyperplasia of all the lymphoid elements, some cellular necrosis and phagocytosis of pigment and of trypanosomes. In the kidneys of the smaller animals ecchymoses are uncommon, and the glomeruli are not involved. In monkeys and the racoon there is a very striking acute interstitial nephritis. The skin lesions and those of the iris show microscopic changes of the type characteristic of trypanosomiasis.

- (19) ROUDSKY (D.). Sur l'Immunité croisée entre le *Trypanosoma lewisi* et le *Tr. duttoni* renforcé. [Cross-Immunity between *T. lewisi* and *T. duttoni*.]—*Compt. Rend. Soc. Biol.* 1912. April 26. Vol. 72. No. 14. pp. 609-611.

Although there are no distinct morphological differences between the *Trypanosoma lewisi* and the *T. duttoni* it is generally agreed that the two are distinct species, the *T. lewisi* being peculiar to the rat and the *T. duttoni* to the mouse.

The author has shown that each parasite can be acclimatised to the other host and this led him to investigate the question of cross immunity, or in other words whether a rat recovered from infection with the *T. lewisi* and hyperimmunised against that parasite would resist infection with the *T. duttoni*, and *vice versa*.

Two experiments were made, and the results indicated that an animal that was immune to the *T. lewisi* was also immune to the *T. duttoni* and *vice versa*. The immunity does not appear to be entirely phagocytory, although there is active phagocytosis. The destruction of the trypanosomes is not localised at the point of inoculation, and it proceeds so slowly that parasites injected into the peritoneum are able to multiply. Their passage into the blood stream is followed by extremely rapid destruction, and for this reason their presence there is quite the exception.

The *T. duttoni* does not appear to constitute a species distinct from the *T. lewisi*, which it closely resembles morphologically. The relative natural immunity possessed by the rat against this parasite is probably only one of chemical adaptation.

- (20) LEGER (A.) & RINGENBACH (J.). Sur la Spécificité de la Propriété trypanolytique des Sérums des Animaux trypanosomés. (Deuxième note.) [The Specific Nature of the Trypanolytic Property of the Serum of Animals infected with Trypanosomiasis.]—*Compt. Rend. Soc. Biol.* 1912. Feb. 23. Vol. 72. No. 7. pp. 267-269.

In a previous note the authors have shown that the serum of animals affected with nagana and surra reacts not only with the homologous trypanosomes, but also with other trypanosomes that are considered to be allied to them, and that with others there is no reaction whatever.

In this new series of investigations experiments have been made with the *T. equinum*, *T. gambiense*, and *T. congolense*.

The technique of the experiments is as follows: The sera have been obtained from guinea-pigs and the viruses from infected mice. To 5 drops of freshly drawn serum are added one drop of citrate solution and one drop of blood from an infected mouse, the mixture being incubated for four hours.

The results obtained were similar to those previously obtained with the parasites of nagana and surra. It was found that the serum of an animal infected with the *T. gambiense* reacted with *T. rhodesiense*, and that of an animal infected with the *T. congolense* with the *T. dimorphon*.

It was observed that the serum of a guinea-pig infected with the *T. pecaudi* was not trypanolytic for any heterologous trypanosome. Although the *T. equinum* differs morphologically from the *T. evansi* the experiments revealed a relationship existing between the two.

This result agrees with the observations originated by WERBITZKI, who has demonstrated the possibility of obtaining experimentally a transmission from a centrosomic to an acentrosomic variety of trypanosome.

In support of their view as to the value of their method of procedure the authors state that the serum of animals infected with the *T. brucei* (Zululand) at the Pasteur Institute proved trypanolytic for the same virus preserved at the Liverpool School of Tropical Medicine.

MISCELLANEOUS.

- (21) NATTAN-LARRIER (L.). La Coloration des Leishmania dans les Coupes. [The Staining of Leishmania in Sections.]—*Compt. Rend. Soc. Biol.* 1912. March 22. Vol. 72. No. 11. pp. 436-438.

The pieces of tissue should not exceed 8 by 4 by 3 mm. They may be fixed in any of the following ways: Acetic sublimate or saturated solution of sublimate, which are probably the best. Alcohol of increasing strength, commencing with 70 per cent. and finishing with 90 per cent. Alcohol fixation may be modified by immersing the pieces of tissue in 2 per cent. formol for three or four hours beforehand.

The following three methods of staining have been employed by the author.

Carbol thionin.—The specimen is stained with carbol thionin for half an hour. It is then washed in distilled water, dehydrated with absolute alcohol, differentiated slowly with clove oil followed by absolute alcohol, and finally clarified with xylol.

The nucleus and centrosome of the Leishmania are stained dark blue, and stand out in contrast to the protoplasm which is faintly tinted blue. Parasites which are extracellular have their outlines well stained.

Kernschwarz and carbol thionin.—Stain with Kernschwarz for a quarter of an hour. Wash thoroughly in distilled water. Stain for half an hour with carbol thionin wash and dehydrate. Differentiate with oil of cloves followed by absolute alcohol until

only the nuclei remain stained. The nucleus and centrosome are stained a greenish-grey, the cytoplasm blue. The protoplasm of the host cells is greyish.

Alum carmine and carbol thionin.—Stain for 24 hours with alum carmine. Wash in distilled water and stain for half an hour with carbol thionin. Differentiate with oil of cloves until the section acquires a reddish-violet fluorescence, and a rapid inspection under the microscope shows that the protoplasm is pink. Dehydrate rapidly in absolute alcohol and clarify in xylol.

- (22) HINDLE (E.). Attempts to transmit "Fowl Pest" [Fowl Plague] by *Argas persicus*.—*Bull. Soc. Path. Exot.* 1912. March. Vol. 5. No. 3. pp. 165-167.

Ticks fed upon infected birds at the height of the disease were unable to infect fresh birds by biting. The virus persists in the digestive tube of the tick for 9 days, but does not pass through the wall. It is not found in the coelom, nor in the coxal liquid.

The intestinal contents are not infective after 14 days. The ticks were kept at 22-28° C.

- (23) CAROUGEAU. Étude générale de l'Osteomalacie chez le Cheval, particulièrement en Madagascar. [Osteomalacia in the Horse, and especially as observed in Madagascar].—*Rev. Gén. de Méd. Vétér.* 1912. Jan. 1. Vol. 19. No. 217. pp. 1-19; and Jan. 15. No. 218. pp. 65-92.

Osteomalacia is in the opinion of the author an infectious disease and is characterised essentially by a progressive demineralisation of the bones.

Clinically the disease is recognised by lameness, pathognomic swelling of the bones of the head and jaws, detachment of ligaments and tendons from the bones, fractures, and frequently considerable wasting.

The lesions, which principally involve the osseous tissue and the bone marrow, are in the nature of a generalised osteomyelitis.

The author states that the disease termed osteomalacia occurring in the horse is entirely different from the conditions grouped under the same name occurring in other species of animals. The name is a misnomer since in the horse death occurs before the bones have become really soft.

The author gives a brief review of the literature on the subject and also mentions the occurrence of the disease in different parts of the world which in temperate countries is sporadic, and in hot countries endemic and epidemic.

The disease appears to have been observed in South Africa for the first time in 1885, and since then a number of observations have been made. Johannesburg appears to be a centre of the disease. The author has not been able to find any records of the disease occurring in Madagascar previous to the conquest, but after the introduction of horses and mules from different parts of the world in 1895 and 1897 the disease made its appearance. It would appear that the Madagascan horses were immune.

The author has seen cases in animals of all ages, the majority occurring in four- or five-year-olds. Cases occur at all altitudes,

in males and females, animals well-fed and ill-fed, and in animals at grass or in the stable.

Animals have been followed for periods of years, but the author has not been able to convince himself that the disease is hereditary. Animals that are used too young are more likely to be attacked than others, and certain races are less resistant than others.

The disease appears to persist in certain places, and this suggests the possibility that it is contagious.

The author's investigations have been directed towards verifying the contagious nature of the disease, studying the pathological anatomy of the disease, and attempting treatment.

In his search for the causal organism the author has not arrived at any definite result. Material has been taken from the bone marrow, synovia, and blood, and a number of ordinary and special media have been employed in attempts to cultivate it but without result.

A variety of organisms have been cultivated from such materials taken with aseptic precautions from animals either living or just dead. The majority of the cultures remained sterile or yielded a small micrococcus which appears "special" to Carougeau.

This organism is very small and frequently occurs in the form of diplococci. Cultures are very meagre, there being a scarcely perceptible growth on agar and only a faint opalescence in broth.

Abundant growths have never been obtained and subcultures have always proved sterile.

Microscopic examination of various materials has not yielded any interesting results.

Blood and materials derived from lesions have been used for inoculation experiments.

1. Horse.—Horses have been inoculated with blood, bone marrow, and bone ground up with sterile broth and filtered, but the results have been negative in every case.

Contact experiments have also failed to transmit the disease to healthy horses.

One experiment was made to transmit the disease to an ox by inoculating it subcutaneously with bone marrow. This also failed. Similarly, experiments with goats, sheep, dogs, rabbits, guinea-pigs, and, with one possible exception, pigs have also proved negative. The author's opinion that the disease is infectious is based upon the following considerations:

The occurrence of the disease under a variety of conditions of feeding, climate, and geological formations compels one to suppose that none of these conditions has a preponderating influence on the causation of the disease. On the other hand the repeated occurrence of the disease in certain places and certain stables suggests that it is of an infective nature. This view is supported by the nature of the lesions, which are those of a chronic irritation.

There is no doubt that defective feeding, inattention to hygiene and so on are contributing causes owing to the decrease of resistance occasioned, but in the author's view there is some essential factor.

Examination of the lesions shows that there is every evidence of active irritation; the bones are not simply decalcified. The inflammatory process is evidenced by the resorption of layers of osseous tissue, the formation of osteoid tissue, the production of a large amount of vascular connective tissue, and finally the nature of lesions in the marrow.

In the early stages it is the bone marrow that is principally involved; the disease is therefore an osteomyelitis which subsequently extends to the osseous tissue and joints.

The theory of infection also explains the digestive disorders always met with in cases of osteomalacia. There is always an unusually excessive elimination of phosphoric acid and calcium, but the quantity eliminated varies from time to time during the course of the disease, being generally greater in the early stages and in many cases nil in the last stages.

The author is of opinion that osteomalacia and rickets are two distinct conditions in the horse.

The lesions involve the bones and the joints and are in the nature of a generalised rarifying ostitis. All the other lesions found in cases of the disease are due to anaemia or general cachexia; there are never generalised lesions. Lesions never occur in the internal organs, save such as are of a secondary nature.

Any of the bones may be affected and the nature of the change is the same in all cases. The bones become lighter, and swollen, and lose their hard compact structure, the bone being converted into a porous sponge-like material which can be easily broken with the fingers. The long bones are less seriously affected than the others, and it is the bones of the head and the jaws that show the most marked alterations.

Joint lesions are constant and are generally more severe in the upper joints of the limbs than the lower ones.

Fractures occur commonly.

The medullary canal is always enlarged and the marrow has a pronounced red colour, although at the centre of the shaft of long bones it may be yellow. Even here there are often haemorrhages. In some cases the marrow resembles spleen pulp. The spaces in the cancellous tissue are enlarged.

Analysis of the bones shows that there is a diminution in the proportion of mineral matter and an increase in the animal matter, the variation depending upon the stage of the disease.

Microscopically the lesion is found to be the replacement of bony tissue by vascular connective tissue. This process may occur in any of the bones, but it is more marked in bones of fibrous origin than in those of cartilaginous origin.

The periosteum is found to be thickened, and beneath this the bone tissue becomes more and more rarified the deeper one examines it from the surface.

Lesions occur in the bone marrow in every case and at all stages of the disease. There is an absence of fat-containing cells and a multiplication of the medullary cells associated with extravasation of blood. Masses of red or yellowish blood pigment may be seen at places. In the later stages there is always some formation of fibrous tissue.

Previous to the deformation of the bones the symptoms presented are those that are to be expected in a disease of this kind.

As already mentioned, fractures commonly occur and it is not uncommon for ligaments and tendons to become detached from the bones even in the early stages of the disease.

Examination of the urine shows that the excretion of phosphoric acid is excessive. Whereas the urine of the horse normally contains about 40-80 centigrammes of phosphoric acid per day the author has found as much as 12.5 grammes per litre in a case of osteomalacia. The quantity excreted varies from case to case and from day to day in the same case.

The alkalinity of the blood is decreased, but in the case of recovery this returns to the normal. The number of red corpuscles may fall as low as 2,500,000 and there is also a decrease in the amount of haemoglobin. The leucocytes increase in number, but the differential counts vary greatly.

The course of the disease is slowly progressive, although at times there may be exacerbations which suddenly render an animal unfit for any work. Recovery is possible even when there are advanced lesions, but such recoveries are exceptional.

Treatment.—Attention to hygiene is of great value in the treatment of the disease. Animals should be left at rest at pasture.

Adrenalin appears to have a beneficial effect, but it proves too costly to be used very extensively.

Lime and magnesium salts have been used with good effect, as also have arsenic and mercury, the latter being administered intramuscularly in the form of calomel.

The author suggests that anhydro-oxymethylene-diphosphoric acid may prove to be a valuable remedy though he has not actually tried it.

- 24) SIEGEL (J.). Einige ergänzende Bemerkungen zum Nachweis der Cytorrhyclescoccen bei Maul- und Klauenseuche. [Supplementary Observations regarding the *Cytorrhycles cocci* in Foot and Mouth Disease.]—*Berlin. Tierärztl. Wochenschr.* 1912. Jan. 11. Vol. 28. No. 2. pp. 27-29.

The author has been able to find the cocci in smears taken from the heart muscle of four calves. In one case there was severe endocarditis with the formation of lesions on the valves of the right side, and in another very marked myocarditis, involving principally the left side and the apex. The author has seen lesions involving the valves of the heart in a pig inoculated experimentally with a pure culture, the inoculation being intraperitoneal. Pure cultures were obtained from two of the hearts.

SIEGEL's observations show that the infectivity of the blood and the number of cocci present in it run parallel.

Very good results have been obtained in the demonstration of the cocci in sections by staining with carbol-fuchsin, diluted three times with water, for half an hour and then washing in water, dehydrating with alcohol and mounting in balsam. The method is more rapid than that previously advised of staining with haematoxylin and methylene blue.

In sections of an unruptured lesion preserved in formalin immediately after the slaughter of the animal practically every cell of the inflamed connective tissue contains a group of the cocci. These generally lie close to the nucleus and are surrounded by a clear zone. The author retracts his previously expressed view that the cocci are actually in the nucleus.

The cocci vary greatly in size, but the majority are very minute.

- (25) UGANDA PROTECTORATE. *Annual Report of the Veterinary Department for the Year 1911-1912*. [Abstracted from the *Ann. Rept. of Dept. of Agriculture for Year ending March 31, 1912*. pp. 18-28. (Entebbe: Govt. Printers).]

East Coast Fever.—The disease is highly endemic over all the districts to the west of the Nile, with the exception of the northern and central counties of Ankole.

In the Eastern Province East Coast Fever is endemic in the Busoga, Lango, and the greater part of the Mbale Districts.

In the Kumi District it is probable that the disease is not highly endemic, and the north eastern portion of the District is known to be free.

It is probable that Karamoja, which would appear to be one of the most heavily stocked districts in the Protectorate as regards cattle and sheep, is also a non-endemic area.

Rinderpest is still confined to the Eastern and Nile Provinces.

Trypanosomiasis.—Losses during the year have been exceptionally heavy. This has been most marked in Bulamwezi where a mortality of over 50 per cent. is reported in some herds. The responsible trypanosome appears to be the *T. pecorum*.

Cases have been reported of a disease closely resembling black quarter in the symptoms presented and the post-mortem appearances. Two cases were met with in which microfilariæ were present in the blood of cattle.

- (26) TODD (J. L.) & WOLBACH (S. B.). *Parasitic Protozoa from the Gambia*. [Second Report of the Expedition of the Liverpool School of Tropical Medicine to the Gambia, 1911.]—*Jl. of Med. Research*. 1912. June. Vol. 26. No. 2. pp. 195-218.

This paper contains the results obtained by the examination of a small number of Gambian animals for parasitic protozoa, the observations being made as opportunity offered while investigations in connection with human trypanosomiasis were being carried out.

The authors state that possibly some of the parasites described are in the nature of post-mortem invaders, in view of the fact that in many cases the animals or birds suffered from wounds and some time elapsed before the examination was made; every precaution was taken, however, to prevent this post-mortem invasion.

Cattle and Horses.—

Trypanosomes.—In the blood of an Ayrshire bull, which was in a moribund condition when seen, large numbers of trypanosomes were found. There were two distinct types present. The

larger parasites had a free flagellum, and the smaller had not. The larger trypanosomes measured 20-25 microns in length, and the smaller 8-11. In both types the centrosome was in many instances terminal.

Similar trypanosomes were found in the blood of native horses.

Antelope.—

Spirochaetes.—A few spirochaetes were found in a specimen of stained blood. They varied considerably in size. The most numerous were short and somewhat thick, measuring 5-7.5 microns in length, but some were seen even shorter than this. The protoplasm was homogeneous or showed a single small clear area. They tapered towards each end slightly, but terminated bluntly.

The other type of spirochaete found was of about the same length but more slender.

Trypanosomes.—A few trypanosomes were present and the majority of these were of the "tadpole" type, measuring about 13 microns. There was also a larger form present measuring 15.5 microns by 3.7.

Rats inoculated with this parasite did not become infected.

Piroplasms.—Red corpuscles containing piroplasms occurred in almost every field of blood smears made from this animal.

The majority of the parasites were rounded in shape and contained a single mass of chromatin which was usually crescentic in form. Bacillary forms were found, but multiple infection of the same cell and bigeminal forms were rarely encountered.

The parasite was a small one, the round form averaging about 1 micron and the bacillary forms 2-2.5.

Granules of chromatin, which resembled the *Anaplasma marginale*, were found either in or at the periphery of some of the red cells. In the event of this proving to be a new species the name *Theileria hippotrangi* is suggested.

There were no cattle in the forest where this antelope was shot but there were considerable herds at no great distance. None of the local chiefs had ever heard of redwater.

Rats.—

Trypanosomes.—Trypanosomes identical in appearance with the *T. lewisi* were found in the blood of rats caught in widely separated localities.

Birds.—

A leucocytozoon was found in the blood of a vulture.

This parasite measured about 7.5 by 3.7 microns. The cytoplasm was coarsely alveolar and stained deep blue. In many of the parasites there was a vacuole like area extending across one end. The nucleus was a loose mass of granular material lying near the middle point of the parasite, and in a few cases towards one end. No free parasites were observed, and all varieties except the small mononuclears and mast cells were found to be invaded, the majority being in large mononuclears. The name *Leucocyto-gregarina neophrontis* is suggested.

Hornbill.—Trypanosomes of two types were found in the blood of a hornbill.

The larger parasite measured about 40 microns. Its cytoplasm stained deeply and showed distinct myonemes.

The smaller parasite measured 26 microns. Myonemes were not so distinct as in the preceding. The centrosome was large and stained deeply, and was always placed about the centre of a curious prolongation of the posterior end of the body.

Doves.—Trypanosomes were found in the blood of two doves. The parasites were similar in morphology to the larger one occurring in the hornbill, but the centrosome was larger and the parasite measured 43 microns.

Two types of trypanosomes were found in the blood of bush fowl. The parasites were large, measuring 56·6 and 60·7 microns. The shorter of the two was provided with a blunt posterior extremity and the larger with a sharp one.

Trichomonads found in the blood of a bush fowl were probably post-mortem invaders, the shot wounds which killed the bird having penetrated the alimentary canal.

Leucocytozoa.—These parasites were found in the blood of four bush fowls. Most of the parasites were the large "adult" forms, the dark-staining females being about five times as numerous as the lighter coloured males. Small "young" forms were found to be very scanty in every case.

The parasites varied greatly in size, but the average size was 19·21 microns in length by 6·7 in width. The smallest measured 5·2·5 microns.

The commonest form of the parasite with its host cell constituted a spindle-shaped body measuring 30·50 microns in length by 6·13 in width, the cytoplasm of the host-cell forming a kind of sheath. None of the cells containing parasites possessed any haemoglobin, nor were the parasites found in large mononuclear leucocytes.

It was impossible to determine with certainty the nature of the host cell, but the nuclei were like those of small mononuclears. Similar nuclei occur however in erythroblastic myelocytes.

The adult female parasites possessed a cytoplasm staining a deep blue. Its texture was coarsely alveolar. The bodies lying within the female parasites which stained like chromatin consisted of two large masses and an indefinite number of smaller granules. The larger of the two large masses was an oval or irregular mass of granules, and the smaller more compact and stained more deeply.

The line which was described as one of the nuclear bodies of a leucocytozoon in the blood of an African hawk was not present in these preparations.

The texture of the protoplasm of the male parasites was finer than that of the females, and it stained less densely. The larger chromatin mass was larger and less compact than in the female parasites, and the smaller mass was less frequently seen, but its relations were the same. The small forms had a deeply staining cytoplasm and one or more chromatin bodies.

Parasites of the Halteridium type were found in a number of different species and an inspection of the specimens suggested that there may be more than one species of halteridium.

Among the parasites found in the blood of reptiles were trypanosomes, haemocytozoa, and haemogregarines.

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- (96) MITTER (S. N.). Some Entozoa of Indian Elephants; and a Gastrodisc (?) from an Indian Zebu.—*Jl. Comp. Path. and Therap.*, 1912. June. Vol. 25. No. 2. pp. 111-115.

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- (102) LEHMANN. Die Amöben als Krankheitsursachen bei den Haustieren [The Amoebae as the Cause of Disease in the Domesticated Animals].—*Centralbl. f. Bakt.*, Abt. I., Orig., 1912. Vol. 62, No. 7, pp. 589-605.
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- (106) PITTALUGA. Ein neuer Blutparasit der afrikanischen Schildkröte. *Clemmys africana*, *Haemoproteus cojali*. [A new Blood Parasite of the African Tortoise].—*Centralbl. f. Bakt.*, I. Abt., Orig., 1912. May 2. Vol. 63. Nos. 2/3, pp. 241-243.
- (107) SERGENT (EDM.), SERGENT (ET.) and SENEVET (G.). Présence d'*Haemogregarina canis* en Algérie. [The presence of *Haemogregarina canis* in Algeria].—*Bull. Soc. Path. Exot.*, 1912. Jan. Vol. 5. No. 1, p. 16.
- (108) YAKIMOFF (W. L.), STOLNIKOFF (W. J.) and KOHL-YAKIMOFF (NINA). Un Hémoparasite nouveau des Chauves-souris. [A new Blood Parasite of the Bat].—*Centralbl. f. Bakt.*, I. Abt., Orig., 1912. Feb. 20. Vol. 62. No. 3/4, pp. 283-287. 1 plate.

Ticks.

- (109) EYSELL (ADOLF). Beiträge zur Biologie der Zecken. [Contribution to the Biology of Ticks].—*Arch. f. Schiffs- und Trop. Hyg.*, 1912, April. Vol. 16. No. 7, pp. 205-212.
- (110) NUTTALL (G. H. F.). Notes on Ticks. II. (i) New Species (*Amblyomma*, *Haemaphysalis*). (ii) *Ixodes putus*: Description of the hitherto unknown Larval Stage.—*Parasitology*, 1912. Feb. Vol. 5. No. 1, pp. 50-60.

Ticks—*continued*.

- (111) WARBURTON (C.). Notes on the Genus *Rhipicephalus*, with the Description of New Species and the Consideration of some Species hitherto described.—*Parasitology*, 1912. Feb. Vol. 5. No. 1. pp. 1-20.
- (112) YAKIMOFF (W. L.), WINOGRADOFF (A. A.) and KOHL-YAKIMOFF (NINA). *Argas persicus persicus* Fischer-Waldheim en Russie d'Europe. [*Argas persicus* in Russia in Europe].—*Bull. Soc. Path. Exot.*, 1912. Jan. Vol. 5. No. 1, p. 39-41.

Unclassed.

- (113) ZOLLENKOPF. Über eine Hühnererkrankung in Graslande Kameruns. [A Disease of Fowls in the Cameroons].—*Arch. f. Schiffs- und Trop. Hyg.*, 1912. March. Vol. 16. No. 6, p. 195.
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ANAPLASMOSIS.

- (114) BEVAN (H. E. W.). *Anaplasmosis of Sheep*.—*Veterinary Jl.* 1912. July. Vol. 68. No. 445. pp. 400-401.

Several outbreaks of disease occurred among sheep in March of this year, the most noticeable symptom in every case being a dropsical condition of the throat. In some cases this has been the only symptom mentioned, but in others the dropsy has been more severe and extensive.

In one outbreak investigated by the author parasites were found in the red blood corpuscles resembling the *Anaplasma* of cattle. In addition, all the changes usually associated with severe anaemia were observed in the blood, the number of cells having fallen to nearly half the normal.

It would appear that the disease has a wide distribution throughout Rhodesia.

- (115) CARPANO (M.). *L'Anaplasmosi nei Bovini della Campagna Romana. (Nota Preventiva)*. [*Anaplasmosis in the Roman Campagna. Preliminary Note.*].—*Il Moderno Zooiatro (Parte Scientifica)*. 1912. Aug. 31. Vol. 23. No. 8. pp. 336-342. With 1 text-figure.

The case was observed by GABBUTI who suspected piroplasmosis, but since the train of symptoms usually observed in cases of *bigeminum* infection was not present blood examinations were made by the author. The animal's temperature at the time of the blood examination was 40.4° C.

GABBUTI gave the author details of six similar cases that he had observed in the same district the previous year. Of these cases five terminated fatally. The disease was met with principally in well-bred imported animals and especially in cows near to calving.

Clinical characters of the disease.—The onset of the disease is marked by a rise of temperature which persists with exacerbations during the whole course of the attack. The appetite is capricious, and rumination may be entirely suspended. The faeces are

normal in consistence but contain a large quantity of mucus. The urine is normal in tint. The conjunctival, buccal, and vaginal mucous membranes are pale and anaemic, but no petechiae or extravasations have been observed.

In the fatal cases death occurred about the 12th to 15th day, and a few days before death there was haemoglobinuria.

In non-fatal cases the temperature falls within two or three days to normal with a few slight exacerbations.

Transmission of the disease.—Since specimens of the *Rhipicephalus bursa* were found upon the cow it appears to be possible that that is the transmitting tick.

Examination of the blood.—A blood count showed that there were about three million erythrocytes per cubic millimetre and about eight thousand leucocytes.

Microscopic examination was made of the blood in fresh and in stained preparations.

Fresh preparations.—In unstained fresh preparations the small size of the parasites made accurate examination difficult. They appeared as somewhat refractile points at the periphery of the red corpuscles. The diplococcus forms appeared to show a slow rotary movement.

Stained preparations.—The best results were obtained with specimens fixed in sublimate alcohol and subjected to prolonged staining with Giemsa (1 in 40), with subsequent decolourisation.

Anaplasma is ordinarily a parasite of the red corpuscles but it may be found free in the plasma. In the majority of cases the parasite is disposed towards the periphery of the host cell, but it may be observed in the centre.

In well differentiated specimens each parasite shows a large chromatin nucleus staining intensely, and a small amount of cytoplasm. The nucleus may be centrally or peripherally placed, and is variable in shape. The parasites vary in size from 0.5 to 1.5 microns in diameter and they may be rounded or oval in shape.

In the diplococcus forms there may be a large surface of contact, the contact may be tangential, or the two parts may be quite separate. In such cases one parasite is a little smaller than the other. These forms are no doubt stages in the multiplication of the organism. One, two, three and not rarely four parasites have been observed in a single corpuscle.

The number of red corpuscles invaded by parasites is variable, but in rich specimens 30 per cent. have been found.

Neither *Piroplasma bigeminum* nor blood parasites other than Anaplasma have been found, although numerous specimens have been carefully examined.

Very severe blood lesions are described. In addition to the destruction of red corpuscles the following abnormalities have been discovered: poikilocytosis, the presence of megalocytes, numerous corpuscles showing punctate basophilia, polychromatophilia, and occasional normoblasts.

Conclusions.—

1. There exists in Italy (Roman Campagna) a disease of cattle caused by a particular protozoal parasite—*Anaplasma marginale*.
2. The parasite gives rise to a severe disease characterised by a marked anaemia with important alterations in the blood corpuscles.
3. The infection is apparently transmitted by the *Rhipicephalus bursa*.

(116) KOIDZUMI (M.). On the Nature of the “Marginal Points” occurring in the Blood Corpuscles of Cattle.—*Centralbl. f. Bakt.*

1. Abt., Orig. 1912. July 17. Vol. 65. No. 4-5. pp. 337-340. With 1 plate.

After a preliminary summary of the views expressed by various authors as to the nature of “marginal points” the author passes on to consider the nature of the parasite as it occurs in the blood of cattle in Formosa. It is stated that there is in Formosa an endemic disease of cattle closely allied to Texas fever. Apparently healthy cattle harbour the virus and imported animals are the principal sufferers.

The author states that as a result of his examinations he has been able to collect strong evidence that marginal points are in reality a stage of *Babesia*, that they persist in the blood of recovered animals for a long time, and serve as the source of infection of imported animals.

Parasites indistinguishable from *Babesia bigemina* appear in the blood of animals suffering from the disease; and when such animals are recovering the marginal points make their appearance in the blood, the large parasites gradually disappearing.

The author states that he has been able to trace several forms intermediate between the two types of the organism. In some cases the parasite assumes the shape of a long-necked flask, the whole of the nuclear substance and the greater part of the cytoplasm being in the rounded part. In others the rounded portion is reduced in size and more dense, the cytoplasm forming merely a tail-like appendage. In still other forms this appendage is reduced to a mere projection.

In the author's opinion the occurrence of what he describes as “large normal forms” and marginal points in the blood at the same time furnishes good reason for believing that the forms above described are intermediate forms.

Marginal points are larger when they have just been formed than in the blood of recovered cattle, and sometimes contain inner structures. Some show a deeply-stained peripheral part with a faintly stained centre, and in others this faintly stained area contains a central intensely stained mass.

Judging from the description given by SIEBER the author thinks that *Anaplasma marginale* as it occurs in Africa is different from the parasite observed by him, and he finds himself unable to accept the view that all the “coccus-like bodies” are *Anaplasma*.

The views expressed by the author in this paper appear to have been based solely on the examination of a number of blood-smears prepared by the Government Veterinary Surgeon in the district.

- (117) SCHELLHASE (W.). Eine Beobachtung über das Vorkommen von Marginalpoints (*Anaplasma marginale*) im Blut von Schafen in Deutsch-Ostafrika. [The Occurrence of *Anaplasma marginale* in the Blood of the Sheep in German East Africa.] Berlin. Tierärztl. Wochenschr. 1912. Vol. 28. No. 28. pp. 511-512.

The author appears to have observed the parasite in the blood of sheep on two occasions only, once in 1909 and once in 1911. The earliest symptoms observed in affected sheep were discharges from the eyes and nose. These were followed by gradual wasting and diarrhoea. There was also intermittent fever and a falling-out of the wool. At the post-mortem nothing characteristic was observed save the lesions typical of anaemia. Parasites (*S. contortus*) were frequently found in the abomasum, but in view of the fact that these were also encountered in healthy sheep the author does not consider that the disease was due to them. The parasite was not found in the early stages of the disease. A very small number of extra-corpuseular organisms were observed, and these were generally very like diplococci in appearance. Transmission experiments could not be undertaken.

- (118) THEILER (A.). Übertragung der Anaplasmosis mittels Zecken. [Transmission of Anaplasmosis by means of Ticks.] Zeitschr. f. Infektionskrankh., parasit. Krankh., u. Hyg. d. Haust. 1912. August 24. Vol. 12. No. 2. pp. 105-116.

1. THE TRANSMISSION OF *Babesia bigemina* AND *Anaplasma marginale* BY MEANS OF THE LARVAE OF *Boophilus decoloratus* (BLUE TICKS).

Origin of the ticks.—The female ticks were collected from African animals which had constantly been at pasture, and which therefore must have been infected with both *Babesia bigemina* and *Anaplasma marginale*. The larvae were hatched out at the laboratory.

Ox 787.—Imported from England and housed for three months after arrival. This animal had placed upon it about 100 larval ticks. On the 11th and 12th days there was observed an elevation of temperature. Blood examination proved negative. During the following days the temperature was somewhat irregular, but did not go higher than 39.5° C. *Babesia* was first observed in the blood on the 26th day and again on the 48th day, the temperature being somewhat irregular in the interval. On the 75th day there was a typical febrile reaction, the temperature remaining high from the 85th to the 100th day. *Anaplasma* first appeared on the third day after the rise of temperature, about 4.5 per cent. of the corpuscles being invaded. The maximum was reached on the 86th day when the percentage was 15.4. The usual lesions of anaemia were observed. After the disappearance of *Anaplasma* the animal recovered.

Since the usual period of incubation after tick infestation varies from 17 to 25 days the rise of temperature on the 11th day must

be considered as accidental, and if the temperature had not been taken and microscopic examination of the blood made the attack of redwater would have passed unnoticed.

The above experiment shows that a single brood of ticks is capable of transmitting the double infection, but furnishes no evidence as to whether a single tick is capable of harbouring both the parasites. This possibility is, however, not excluded.

2. THE TRANSMISSION OF ANAPLASMA BY MEANS OF LARVAL BLUE TICKS TO ENGLISH CATTLE IMMUNISED AGAINST *Babesia bigemina*.

Experiment A.

Ox 922.—Immunised against redwater in England by a subcutaneous inoculation of 10 cc. of immune redwater blood.

On the 5th day after inoculation the temperature rose to 40·6° C. Blood examination was negative, but the animal was given a dose of trypanblue. The temperature fell rapidly and *Babesia bigemina* was observed for the first time on the 7th day after the initial rise of temperature and again 6 days later. The parasites were present in very small numbers only. The animal was housed directly it arrived in South Africa and observations carried out daily for a long period shewed that its temperature was normal.

The fully-engorged females from which the larvae were obtained were collected from animals in Natal, and since both babesiasis and anaplasmosis occur in Natal it is probable that the animals from which the ticks were obtained were immune to anaplasmosis.

Seventy-seven days after its arrival the animal had placed upon it a large number of larvae. From the 10th to the 14th day there was a slight febrile reaction, *Babesia* being observed in the blood in very small numbers on one occasion only. Subsequently the temperature was somewhat irregular but examination of the blood proved negative.

On the 55th day there was a further reaction which lasted for 18 days. Anaplasma was observed in very small numbers on the 55th day and two days later 8·9 per cent. of the red corpuscles were invaded. By the 61st day the percentage of invaded cells had risen to 18·8, but after this there was a rapid fall in the number, the percentage being only 0·6 four days later. The parasite was still demonstrable in very small numbers on the 75th day, and appeared to be of the "*centrale*" variety. On the 71st day *Spirochaeta theileri* was observed for the first time and from the 76th to the 86th day there was a second rise of temperature, *Anaplasma marginale* var. *centrale* again appearing in the blood with a maximum invasion of the red corpuscles of 5·4 per cent.

It appears to be possible that the first rise of temperature was caused by the spirochaete and that the appearance of *Babesia* in the blood was more of the nature of an accident, such as are frequently seen in immune animals.

Experiment B.

Ox 925.—Immunised against redwater in England as in the previous case. The resulting reaction was very mild, parasites being observed in the blood on a number of occasions, but never in large numbers.

The ticks used in this experiment were of the same brood as those used in the previous experiment.

Several thousand ticks were placed on this animal on the 77th day after its arrival, the animal's temperature having been taken daily and found to be normal during the whole of the period.

The first rise of temperature occurred on the 9th day and lasted for a week. Immediately afterwards there was a second rise which was somewhat irregular and lasted till the 30th day. *Babesia bigemina* was found on the 12th day.

From the 45th day there was a third reaction during which the evening temperature rose as high as 41.0° C. Repeated examinations of the blood were made, but no abnormality save slight anisocytosis was discovered. A fourth reaction commenced on the 114th day during which a few Anaplasmata were found which belonged to the "*centrale*" variety.

Possibly in this case also the irregular temperature reactions were referable to the presence of spirochaetes which were overlooked; the possibility is not however excluded that the severe infestation with ticks was responsible. In this case the appearance of Anaplasma was delayed and would no doubt have escaped observation had not the microscopic examinations been continued for so long.

3. THE TRANSMISSION OF A PURE ANAPLASMA INFECTION TO SUSCEPTIBLE CATTLE BY MEANS OF TICKS (*B. decoloratus*).

Ox 934 was inoculated with blood from a calf that was infected with Anaplasma and *Babesia mutans*. It could be proved in this case that *Babesia bigemina* infection was certainly excluded, as was shown by the typical reactions obtained in both animals when subsequently inoculated with redwater.

The pure Anaplasma infection was obtained by means of blue ticks because it has been proved that this tick is not a host of the *B. mutans*. Ticks free from parasites had to be used in this experiment and these were obtained by collecting the fully engorged females from horses.

The larvae which were hatched out at the laboratory were placed on a clean, freshly imported animal, and if no reaction resulted it might be taken that the ticks from the horses were clean and also that the larvae obtained from uninfected animals must remain clean.

Ox 931 had placed upon it larval blue ticks derived from females that had engorged themselves upon horses. There was no reaction. From this it might be concluded that the ticks obtained from the horse were not infected. The fully engorged females were collected from Ox 931 and the larvae derived from

these were placed on Ox 934 which was infected with *Anaplasma* and *Babesia mutans*. A large number of ticks were used and from 20-30 days later the fully engorged females were collected. The larvae derived from these ticks could only be infected with *Anaplasma*.

Calf 1168, which was born at the laboratory and had been kept free from ticks, had placed upon it a number of young larvae obtained from Ox 934. The larvae went through the stages of development and began to drop off on the 24th day. Repeated examinations of the blood proved negative. On the 52nd day there was a febrile reaction which lasted for 18 days, the temperature rising to 40° C. *Anaplasma* was found in the blood on the day on which the temperature rose. The maximum percentage of corpuscles infected was 7·8 (on the second day of the reaction). From the 8th day to the end of the reaction the number of invaded corpuscles amounted to 1 per cent. The blood lesions of anaemia were pronounced but soon disappeared.

Proof of the purity of the infection.—To furnish proof of the purity of the infection of Calf 1168 blood was drawn from it and injected into Ox 1217 which had arrived about a fortnight previously and which, since arrival, had been housed and kept free of ticks.

A febrile reaction commenced on the 22nd day and lasted till the 31st. *Anaplasma* was found to be present in the blood from the 22nd to the 44th day, there being also pronounced evidence of anaemia. Neither *Babesia bigemina* nor *B. mutans* was found.

The susceptibility of Ox 1217 to *B. bigemina* was tested by inoculating it with blood from Ox 1216 which had been infected with pure redwater. This resulted in a positive reaction and the animal was treated with trypanblue.

Ox 1218. For sixteen days after its arrival this animal was kept free from ticks and its temperature noted daily without any elevations being observed. It then had placed upon it larval ticks derived from females from Ox 934.

On the 22nd day there was a slight rise of temperature, but blood examination proved negative and the rise was obviously of an accidental nature. Commencing on the 70th day there was a slight but typical febrile reaction which lasted for 15 days. *Anaplasma* was observed in the blood throughout the reaction and there was evidence of anaemia.

Proof was furnished in the same manner as in the case of Ox 1217 that the infection contracted by Ox 1218 was pure.

The susceptibility of this animal to *B. bigemina* infection was also proved in a similar manner.

4. TRANSMISSION EXPERIMENTS WITH THE LARVAE OF *Rhipicephalus simus*.

The fully engorged female ticks were obtained from Natal, and it was presumed that the larvae derived from them were infected either with *Babesia* or *Anaplasma*. The ox is not the most favour-

able host for *Rhipicephalus simus* larvae as only a few attach themselves to this animal.

Ox 930.—This animal was housed for a period of three and a half months after its arrival and its temperature noted daily. It then had placed on it a number of *simus* larvae. On the following day it was observed that only a small number had attached themselves.

On the 75th day there was a rise of temperature which lasted until the 100th day, the febrile reaction running a typical course. Anaplasma appeared in the blood at the commencement of the reaction and persisted throughout it. Lesions of anaemia were observed in the blood.

The purity of the infection in this case was tested by using 50 cc. of the blood taken some six months after the animal's recovery and injecting it into a susceptible English animal (1213).

The resulting reaction was a very severe one, the animal suffering from marked anaemia complicated by loss of appetite, inability to stand, and acceleration of respiration. Treatment was resorted to and the animal recovered. Babesia was not observed in the blood during the reaction, but Anaplasma was present in very large numbers.

The susceptibility of Ox 930 to babesiasis was tested by placing on it infected larval blue ticks derived from females engorged on an immune animal. There was a positive reaction, *Babesia bigemina* appearing in the blood from the 29th day. There was a rise of temperature on the 20th day and *Spirochaeta theileri* was found to be present in the blood.

Conclusions.

Five imported English animals and one stall-born Africander calf were used for the experimental transmission of Anaplasma by means of ticks. All the animals were susceptible to the disease. Two of the English animals were immune to redwater the immunity having been conferred experimentally before exportation.

Anaplasmosis was transmitted in every case. In the first experiment both anaplasmosis and babesiasis were transmitted by means of larval blue ticks derived from females engorged on animals immune to redwater and anaplasmosis. In the second experiment the ticks transmitted anaplasmosis to animals immune to redwater. In the third experiment the ticks used were originally obtained from horses and were proved to be entirely free from infection. These ticks were infected by placing them on an animal which had passed through anaplasmosis and *mutans* babesiasis. Only the Anaplasma infection was transmitted as shewn by subsequent inoculations of the English animals with blood. In the fourth experiment it was incidentally proved that the larvae of *Rhipicephalus simus* are capable of transmitting a pure Anaplasma infection. All the animals which passed through an attack of pure anaplasmosis remained susceptible to infection with *Babesia bigemina*.

It must be considered as proved that Anaplasma can be transmitted by means of ticks either by itself or together with *B. bigemina* and *Spirochaeta theileri*. This proves that anaplasmosis and babesiasis are independent conditions. Reference must be made to the comparatively long period of incubation after tick infestation. Full use was made of this fact in the protective inoculation against anaplasmosis (*A. marginale* var. *centrale* against *A. marginale* infection).

ADDENDUM.

The Virus of Anaplasmosis does not pass through a Berkefeld Filter.—In all his investigations the author has considered the rounded bodies contained in the red corpuscles, either centrally or peripherally placed, to be of a parasitic and protozoal nature. This view is based on the staining and biological characters of the bodies.

In spite of the proof furnished in support of this view the idea suggested itself that the so-called *Anaplasma* might not be parasitic in nature but only a phenomenon associated with the disease—a cell-inclusion such as have been demonstrated in other diseases. If this were the case the virus would be invisible. Since the disease is transmissible by inoculation an answer to the question could be obtained by filtration experiments. With this object the following experiment was undertaken.

A quantity of blood was taken from Ox 1192 which had contracted anaplasmosis naturally and in which 50 per cent. of the corpuscles were invaded. This was diluted with 12 volumes of salt solution and passed through a Nordmeyer-Berkefeld filter. Three hundred cubic centimetres of the filtrate were inoculated subcutaneously into Ox 1211 (English). There was no febrile reaction, and parasites did not appear in the blood.

Six weeks later the animal was tested with regard to its immunity to redwater and anaplasmosis. It was given 5 cc. of blood from each of the Oxen 1216 and 1212 which were immune to redwater and anaplasmosis.

A febrile reaction resulted and *B. bigemina* was found in the blood on the 7th day. Treatment with trypanblue was resorted to and the parasites disappeared. From the 50th to the 65th day *Anaplasma* was demonstrable in the blood. The parasite was of the “*centrale*” variety. The reaction was slight.

BABESIASIS.

- (119) PÉCAUD (G.). *La Piroplasmose Bovine au Dahomey*. [Bovine Piroplasmosis in Dahomey].—*Bull. Soc. Path. Exot.* 1912. July. Vol. 5. No. 7. pp. 482-486.

Young animals are principally attacked and in them the disease is benign. Among adult animals only a small proportion are affected, but the disease is far more serious. The principal symptoms are: Weakness, anaemia and cachexia associated with slight inco-ordination, more or less elevation of temperature, diarrhoea, and slight jaundice. Haemoglobinuria is only exceptionally observed. Death takes place in severe cases in fifteen to twenty days.

The lesions found at the post-mortem are not marked. There may be oedema of the lungs with extensive areas of congestion, a large quantity of liquid in the pericardium, petechiae on the heart. The spleen may be enlarged and congested, and in two cases was found to be ruptured. There is enlargement and congestion of the liver. The kidneys may show haemorrhagic

nephritis. The blood is watery and the number of red corpuscles greatly reduced.

The parasite occurs in the blood in four well-marked forms :

(a) Classical bigeminal forms, with, rarely, four parasites in a single corpuscle.

(b) Ring forms having the chromatin collected at one point and a central vacuole.

(c) Rod-shaped and comma-shaped parasites half the substance of which is composed of chromatin.

(d) Small forms arranged in the form of a Maltese cross. The parasites are most abundant at the commencement of and during the acute stage of the disease. They do not disappear completely from the blood, and may reappear if the vitality of the animal is lowered from some other cause.

Details are given of a fatal case which was complicated with ulcerative stomatitis and gastritis.

The following is a summary of a benign case in a calf four and a half months old.

The first symptom observed was weakness of the quarters and slight greenish diarrhoea associated with a little elevation of temperature. For the following five days the condition remained the same, twin piroplasms being very scanty in the blood. Two days later the temperature had risen to 39° C., parasites were numerous in the blood, particularly the twin and ring forms. Punctate basophilia was also observed. The following day piroplasms were rather less numerous. The symptoms gradually abated, piroplasms disappearing from the blood, and the calf appeared to have made a complete recovery two months later. Shortly afterwards the animal was used for the preparation of Jenner's vaccine, and during the period of eruption and for some days after a small number of twin parasites were discoverable in the blood.

Five cubic centimetres of the blood of this calf, withdrawn when the piroplasms were not scanty, were injected into a calf one and a half months old. On the ninth and tenth days the animal appeared to be ill and refused food. There was slight fever. Parasites first appeared in the blood on the nineteenth day, a few twin forms being observed. About a week later ring and rod forms were found. Within ten days the parasites had disappeared and the animal recovered rapidly.

Inoculation experiments have been repeated on two occasions with positive results, every care being taken to avoid the possibility of the results being due to natural infections.

Ticks are very numerous in Dahomey at the commencement of the rains, and the species found are *B. annulatus*, *B. decoloratus* and unidentified species of *Amblyomma* and *Rhipicephalus*. Piroplasmosis appears after the rains only (November and December).

Injections of twenty-five to fifty centigrammes of atoxyl cause a rapid decrease in the number of parasites.

THEILERIASIS.

- (120) THEILER (A.). Weitere Beobachtungen, betreffend die Übertragung von Küstenfieber vermittels Zecken. [Further Observations regarding the Transmission of East Coast Fever by means of Ticks.]—*Zeitschr. f. Infektionskrankh. Parasit. Krankh. u. Hyg. d. Haust.* 1912. Vol. 12. No. 1. pp. 26-42.

As is well known the parasite of East Coast fever can be transmitted by five species of ticks, viz., *Rhipicephalus appendiculatus*, *R. evertsi*, *R. simus*, *R. nitens*, and *R. capensis*.

The species which occurs most frequently in countries where the disease is known is the *R. appendiculatus*, and it was with this tick that the author's experiments were carried out.

EXPERIMENT 1.

- A. *Brown ticks which have been infected with East Coast fever as larvae, transmit the disease to susceptible animals in the nymph stage, but are non-infective in the adult stage.*

R. appendiculatus Imago (No. 268).

Fully engorged female ticks, obtained from Natal, deposited eggs a few days after reception, and these hatched out a month later.

Larvae placed on an infected animal for three days when the parasites were numerous in the blood.

Larvae moulted normally and the resulting nymphs were placed on a susceptible animal. The disease developed on the 15th day, and 12 days later the animal was slaughtered for inoculation experiments. The nymphs engorged themselves, and after moulting were placed upon two other animals.

(b) *R. appendiculatus* Imago (No. 309).

Larvae derived from fully engorged females were placed on calf 700. They engorged themselves and dropped off at a time when the parasites were numerous in the blood. As nymphs they were placed on calf 917 and the animal developed the disease and died; parasites being numerous in the blood and agamonts and gamonts in the glands. The females developed from nymphs collected from this calf were placed upon two other animals (1145 and 1088), in order to test their infectivity.

(c) *R. appendiculatus* (No. 335).

Larvae placed upon calf 917 when suffering from the disease, the ticks being collected at a time when there were numerous parasites in the blood.

The resulting nymphs were used together with those from animals 700 and 923 (ticks 268 and 309) on eight animals (Nos. 561, 908, 919, 1011, 914, 1012, 1026, and 1040).

Six of the eight animals contracted the disease.

- B. *Brown ticks which have been infected with East Coast fever as larvae are uninformative for animals that are immune to the disease as a result of inoculation, and are cleansed by being placed upon such animals.*

Larvae Nos. 309, 268, and 335 were fed upon infected animals and proved to be uninformative in their nymphal stage by placing them upon susceptible animals, but nymphs derived from exactly the same source proved to be non-informative when placed upon 10 animals that had been rendered immune to the disease experimentally.

- C. *Ticks which have passed their nymphal stage either upon susceptible animals to which they have transmitted the disease or upon immunised animals fail to transmit the disease to susceptible animals in the adult stage.*

Six experiments of this nature were carried out with negative results.

The author draws the following conclusions from this series of experiments:—

Ticks which fed as larvae on diseased animals and transmitted the disease as nymphs to eight susceptible animals, or which fed as nymphs on twelve immunised animals, failed to transmit the disease when placed in larger numbers on six susceptible animals.

A number of animals were used in the experiment and a large number of ticks were employed so that the possibility of any accident can be excluded.

EXPERIMENT 2.

Ticks which have fed upon animals suffering from East Coast fever do not always transmit the disease in the succeeding stage.

Details regarding the ticks used.

1. *R. appendiculatus* No. 363.

Larvae placed upon ox 1013 on the 7th day after the rise of temperature, and collected eight days later; this being three days before the animal died from the disease. Parasites were numerous in the blood and all the developmental stages of the parasite were discovered in the glands.

2. *R. appendiculatus* No. 364.

These ticks were placed upon an infected animal (No. 908) on the 5th day of the disease and fed for 10 days.

3. *R. appendiculatus* No. 355.

The larvae fed for 5-7 days upon an infected animal (No. 913).

4. *R. appendiculatus* No. 356.

Fed for 3-6 days upon animal 914. This animal recovered from the disease.

5. *R. appendiculatus* No. 349.

Fully engorged nymphs were taken from a diseased animal in Natal and moulted in the laboratory.

6. *R. appendiculatus* No. 411.

Fed both as larvae and as nymphs on affected animals (Nos. 914 and 1053).

7. *R. appendiculatus* No. 426.

Larvae hatched out from eggs laid by engorged females obtained from Natal. Fed upon a susceptible animal and after moulting were fed upon Ox 1111 which was suffering from East Coast fever.

8. *R. appendiculatus* No. 373.

The mature females were obtained from animals 906, 1088, 1009, and 1021. The larvae were placed on ox 909 when it was the subject of the disease.

TESTS OF THE INFECTIVITY OF THE TICKS.

(a) *Brown nymphs* 363, 364, 355, 356, and 349.

1. Ox 627.—This animal had previously been used for inoculation experiments in connection with the transmission of the disease with negative results.

On three separate occasions this animal had placed on it a number of nymphs (Nos. 363, 364, 355, and 356), but in no case was there any reaction.

Subsequently there were placed upon it 6 imagines obtained from Natal. Three of these attached themselves. The animal developed the disease which terminated fatally.

2. Ox 911.—This animal had also been used in transmission experiments with negative results.

On two occasions this animal had placed on it 20 nymphs (Nos. 364, 355 and 356) without there being any reaction.

Six brown ticks from Natal were subsequently placed upon it with the result that a fatal attack of the disease was set up.

3. Ox 1014.—The history of this animal was the same as of the preceding ones save that nymphs failed to set up the disease on four separate occasions.

4. Ox 1068.—History like that of No. 1.

(b) *Brown Nymphs* Nos. 363 and 373.

5. Ox 1037.—Had been used in experimental work without result. Twenty-seven ticks (No. 363) failed to cause any reaction. Five months later 20 nymphs from ox 909 (No. 373) caused a fatal reaction.

(c) *Brown Nymphs* Nos. 363, 364, 349, 411, and 426.

6. Ox 1046.—Animal had been used unsuccessfully in transmission experiments.

On two occasions nymphs and on one occasion adults from Natal were placed upon this animal without result.

Subsequently a fatal attack of the disease was set up by 10 adults, 8 (No. 411) from animal 1053, and 2 (No. 426) from ex 1111. Only five of the ticks actually attached themselves.

(d) *Brown Nymphs* Nos. 363 and 364.

7. Ox 1043.—Previous history as before.

First batch of ticks (nymphs No. 364) failed to set up the disease.

Six weeks later a second batch of the same ticks set up a fatal attack.

8. Ox 1017. As ox 1043, save that the ticks which failed to set up the disease were No. 363, and those which caused the fatal attack were No. 364.

(e) *R. appendiculatus* (Nos. 364, 349, 411, and 426).

9. Ox 1090.—Had not been used before. Was obtained from a district free of the disease.

Ten ticks No. 364 were placed upon the animal but only 6 attached themselves. There was no reaction.

A month later four adult ticks from Natal were placed upon the animal, two attached themselves without producing any reaction.

Three weeks later 6 adults (No. 411) from animal 1053 were used, five of which attached themselves.

A week later two more of the same ticks were put on the animal and also two from animal 1111 (No. 426). The animal contracted the disease and died.

(f) *R. appendiculatus* Nos. 363, 364, and 349.

10. Ox 1082.—This animal was obtained from a district free of the disease and had not been used for experiment.

On two separate occasions brown nymphs (Nos. 363 and 364 respectively) were placed upon this animal without producing a reaction.

Subsequently a reaction which terminated fatally was obtained by placing 6 adult ticks (No. 349) obtained from Natal on the animal.

(g) *R. appendiculatus* nymphs No. 364.

11. Ox 1050.—This animal had been used previously for a transmission experiment which was unsuccessful; infection and death followed the attachment of 7 nymphs (No. 364).

From these experiments it may be seen that;

Ticks No. 363 were not infected.

Not all the ticks in batch 364 were infective.

Batches 355 and 356 were not infected.

Batch No. 349 were infective, but in two cases in which two and three ticks were used the disease was not transmitted.

Batch No. 373 were infective.

Batches 411 and 426 were infective.

EXPERIMENT 3.

Adult brown ticks which fed as larvae on infected animals and were virulent as nymphs failed to transmit the disease when the nymph stage was passed upon rabbits.

1. *R. appendiculatus* No. 342.

The larvae were derived from adults collected in Natal and were placed upon ox 923 at a time when parasites were numerous in its blood. The engorged larvae were collected and moulted normally. A number of them were placed on animal 596 and transmitted the disease, others were placed upon a rabbit, the engorged nymphs being collected.

2. *R. appendiculatus* No. 309.

Larvae derived from adults collected in Natal were placed on calf 700 which was suffering from the disease. After moulting some were placed on calf 917 and transmitted the disease, and others were placed on a rabbit.

TEST OF THE VIRULENCE OF THE TICKS.

1. Ticks from both of the rabbits were placed on ox 1059 in succession without producing the disease. The animal subsequently died from East Coast fever.

2. Ox 1019 had three batches of ticks from the rabbits placed upon it, two from the first and one from the second, without any reaction being caused. The animal subsequently died from the disease naturally contracted.

EXPERIMENT 4.

Adult ticks which have passed their nymph stage on animals which have recovered from East Coast fever are unable to transmit the disease to susceptible animals.

The immune animal used in this experiment was No. 914. This ox was infected by nymphs Nos. 268, 335, and 309. Blue bodies were demonstrable in its glands, and *T. parva* was present in its blood. The animal recovered.

The ticks which were placed upon the immune animal were nymphs No. 298 obtained from ox 868.

TEST OF THE TICKS.

The engorged nymphs were collected and after moulting were placed on ox 1021. There was no reaction.

Calf 1130 also had some of the ticks placed on it but without any subsequent reaction.

CONCLUSIONS.

1. Brown adults which were infected as larvae, and which transmitted the disease as nymphs, are not infective for susceptible animals. Three different batches of ticks were used. In the nymph stage they transmitted the disease to eight animals, but failed to transmit it to two susceptible animals in the adult stage.

2. Ticks belonging to the same batches which had reached the nymph stage on animals experimentally immunised against East Coast fever failed to transmit the disease in six cases in the adult stage. This proves that brown ticks which become infected in one stage are cleansed in the following stage whether they engorge themselves on immune or susceptible animals.

3. Ticks infected as larvae which passed the nymph stage on rabbits were not infective for susceptible animals as adults. This confirms the foregoing fact that a tick loses its infection the first time it feeds on a susceptible or immune animal.

4. Clean or infected ticks which feed on a recovered (immune) animal do not transmit the disease in their next stage. This conclusion was arrived at eight years ago. (See *Annual Report of the Government Veterinary Bacteriologist, Transvaal Dept. of Agriculture*, 1904-1905.)

5. It has been shown that different batches of ticks which were collected at the same time and kept under the same conditions do not transmit the disease during the next stage even when placed upon animals in large numbers. Other batches kept under the same conditions and collected in the same way infect a few animals only, while still others prove to be infective on every occasion, and even when used in small numbers only.

It is difficult to explain this fact but it is quite possible that external circumstances have an influence in some way or other. Those ticks which failed to transmit the disease were fed during the coldest periods of the year. This is possibly the explanation of the fact that the disease in the field is relatively less severe during the winter months; but the fact should not be lost sight of that at this period of the year the ticks are not particularly active.

TRYPANOSOMIASIS.

(121) BLACKLOCK (B.). The Trypanosomes found in a Horse Naturally Infected in the Gambia. A Double Infection.—*Ann. Trop. Med. & Parasit.* 1912. May 29. Vol. 6. No. 1. B. pp. 107-116.

In the first place reference is made to the conclusion of a previous paper* published in 1911, the reference being as follows:—

"A former paper (1911) dealt with the trypanosomes found in two horses naturally infected in the Gambia. These horses were referred to as *Horse A* and *Horse B*. The former, *Horse A*, contained in its peripheral blood two forms of parasite, one long the other short; the latter, *Horse B*, presented one form of parasite only. The conclusion of the paper was as follows:—

"(1) We consider the trypanosome found in *Horse B* to be *T. dimorphon*, *sensu* Laveran and Mesnil.

"(2) The long form in *Horse A* appears to us to be *T. vivax*.

"(3) As regards the short form found in *Horse A*, we do not feel justified at the present stage in assigning its position. It may be a Dimorphon-like trypanosome of low pathogenicity, or simply a modification of the long parasite of *Horse A*."

The two forms of trypanosomes were separated by animal inoculation and from this the author adduces evidence of the dual infection. Apart from the morphological differences the following evidence proves this duality.

The long form of trypanosome has been kept running for a year in goats, maintaining its morphology for the whole of the time,

* YORKE & BLACKLOCK. The Trypanosomes found in Two Horses naturally infected in the Gambia. *Ann. Trop. Med. & Parasit.* 1911. Vol. 5. No. 3. p. 413.

and short forms have never appeared in any goat save in one instance at the first passage. The short form was in the minority in the proportion of about one to a thousand of the long forms. In monkeys, guinea-pigs, rabbits, rats, mice, and dogs only the short form has appeared, save in one rabbit of the first passage (inoculated from the horse) in which long forms predominated till the last day, when the short forms suddenly became numerous. The short form after being carried through a number of smaller animals was inoculated back into a horse. The blood of this animal was found to be highly infective for a goat, dogs, rabbits, guinea-pigs, rats, and mice, but only the short forms appeared.

Many laboratory animals having proved themselves refractory to the long form were readily infected with the short form.

A small number of laboratory animals inoculated intraperitoneally from goats infected with the long form became infected, and parasites were found in their blood for brief periods, but the strain could not be carried on in the same species.

Pathogenicity of T. vivax in goats.—The average period of incubation was nine days, and the period of illness thirty-one days. Every goat inoculated became infected, and none recovered. There was no marked diminution in the period of incubation or in the duration of the disease.

It was observed that *T. vivax* after it had been preserved for some time in goats gradually became slower and slower in its movements. This change of movement commenced in October and persisted till March, when the original motility returned. Circumstances indicated that this slackening of the movement was not due to conditions of temperature in the laboratory.

Two strains of short forms were kept running, one in rats only derived from the horse inoculated from a small laboratory animal, and one in rabbits derived from the original horse. There was observed to be a marked increase in virulence of the strain passed through the horse, the period of incubation being reduced from eighteen to four days.

- (122) BRUCE (Sir D.), HARVEY (D.), HAMERTON (A. E.), DAVEY (J. B.), & LADY BRUCE. *The Morphology of Trypanosoma simiae*, sp. nov.—*Proc. Roy. Soc.* 1912. August 24. Series B. Vol. 85. No. B. 581. pp. 477-481. With 1 plate and 2 text-figures.

This trypanosome is remarkable in that it attacks only such widely different animals as the monkey and the goat. Oxen, baboons, dogs, guinea-pigs and white rats appear to be immune. The trypanosome is fatal with great rapidity to monkeys. In a series of nineteen cases the average duration of life after the trypanosomes were first seen in the blood was only 2·9 days. In the Kasu Hill district the carrier is *Glossina morsitans*.

In the living unstained state the trypanosome shows active translatory movements. In preparations fixed with osmic acid and stained by Giemsa the measurements of the trypanosome were found to be as follows. In the monkey the average length was

18.1 microns, the maximum being 24 and the minimum 14. In the goat the average length of the trypanosome was about one micron less than in the monkey, the maximum being 21 and the minimum 14.

The species is monomorphic. The average breadth is 1.75 microns, the maximum being 2.75 and the minimum 1. The body is frequently extended in a straight line. The posterior extremity is blunted, pointed or rounded, and there is frequently the appearance of a vacuole at the extreme end. The body substance is free from granules. The nucleus is oval and situated about the middle of the body. The micronucleus is small and round, situated about $1\frac{1}{2}$ microns from the posterior end of the body. A peculiarity is that it is always placed at the edge of the trypanosome and appears to protrude a little. The trypanosome differs from the *T. vivax* and *T. uniforme* in that the undulating membrane is well developed. It is difficult to say whether the species has a free flagellum or not.

In the monkey these trypanosomes swarm in enormous numbers in the blood. Large masses of them can sometimes be seen, the masses being sufficiently large to fill up the whole field of the microscope. It would appear as if multiplication took place so rapidly that individual trypanosomes had not time to disengage themselves. Other division forms are observed in which the trypanosomes appear to be joined together by their non-flagellar ends.

"Conclusions. (1). *T. simiae* is a well-defined species, easily separated by its morphology alone from the other trypanosomes which have been described as causing disease among domesticated animals.

"(2) It sets up a chronic disease in goats, but is chiefly remarkable for its rapidly fatal action on monkeys.

"(3) In Nyasaland it is carried by *G. morsitans* and in this district—Central Angoniland—this tsetse-fly is found to be heavily infected with this trypanosome."

(123) KINGHORN (Allan) & YORKE (Warrington). Trypanosomes obtained by feeding Wild *Glossina morsitans* on Monkeys in the Luangwa Valley, Northern Rhodesia.—*Ann. Trop. Med. & Parasit.* 1912. Sept. 12. Vol. 6. No. 3. A. pp. 317-325.

Unfortunately owing to the lack of healthy goats and sheep these animals could not be used in the experiments with wild *G. morsitans*, monkeys being the only animals available for the purpose. In spite of this a certain amount of evidence exists that both *T. vivax* and *T. nanum* are naturally transmitted by this fly, although proof is not forthcoming.

In the first series of experiments 3,008 flies were fed on 28 monkeys. Nineteen of the monkeys became infected. The trypanosomes isolated were the *T. rhodesiense*, *T. pecorum* and a third hitherto undescribed parasite for which the name *T. ignotum* is suggested. The proportion of wild fly infected with these trypanosomes is given as follows: *T. ignotum* 1:300, *T. rhodesiense* 1:534, *T. pecorum* 1:1600.

T. ignotum.—This parasite was obtained in ten experiments.

Morphology.—In fresh preparations this parasite appears as a comparatively short and slender organism. It is fairly actively motile but exhibits no marked degree of translatory power. In stained preparations it is observed that the trypanosome is slender, the posterior extremity is obtuse or bluntly rounded, the nucleus is rounded or oval and lies at the middle of the body. The centrosome is small and while usually situated near the posterior extremity it may be separated from it by an appreciable interval. The undulating membrane is feebly developed and a short free flagellum is only occasionally seen. The cytoplasm shows no granules or vacuoles. The average length of 200 individuals was 17 microns, the maximum 23, and the minimum 12.

Pathogenicity.—The virulence of the trypanosome is very great for monkeys whether the infection be through the agency of flies or by direct transmission from monkey to monkey. The average period of incubation was seven days and death occurred 2 or 3 days after the parasites appeared in the peripheral blood. A rabbit was successfully infected by inoculation from a monkey, the period of incubation was 22 days and the duration of the disease 66. One guinea-pig, five rats, and four mice were found to be refractory and negative results followed the feeding of infective flies upon rats.

Diagnosis.—*T. ignotum* most closely resembles *T. uniforme* but it is easily distinguished from that organism by the absence of a free flagellum and also by differences in the pathogenic range. Although the parasite has been isolated from wild *G. morsitans* more frequently than any other trypanosome it has never been found in game or domesticated stock and no information is at present obtainable regarding its original host.

Transmission.—In one experiment the infective fly was determined to be one of a group of ten. These were then killed and dissected. In nine of them no trypanosomes were found, while in the tenth a heavy infection of the proboscis was encountered. The gut, salivary glands and sucking stomach were negative.

(124) KINGHORN (Allan) & YORKE (Warrington). Trypanosomes infecting Game and Domestic Stock in the Luangwa Valley, North-Eastern Rhodesia.—*Ann. Trop. Med. & Parasit.* 1912. Sept. 12. Vol. 6. No. 3. A. pp. 301-315.

Glossina morsitans is everywhere abundant in this valley, but in spite of that a large and varied selection of game is found, and probably because of that domestic stock is very scarce.

The authors found it advantageous to examine the blood in preparations dry-fixed in alcohol and stained by Giesma rather than to examine moist preparations owing to the rapidity with which the trypanosomes degenerated. The smears were made from blood from one of the arteries immediately after the animals were shot.

Owing to the impossibility of getting "clean" sheep and goats all the inoculations from game were made into monkeys and rats. The authors recommend, however, that when possible sheep and goats should be used as well since they are susceptible to most of

the pathogenic trypanosomes. This is important when dealing with the *T. vivax* and *T. nanum*.

One hundred and twenty-seven head of game, comprising nineteen genera, were examined, and trypanosomes were found by direct examination, by inoculation, or by both methods in thirty-three. Direct examination was successful in twenty-six cases. Had a number of smears been examined from each animal this number might possibly have been higher. In several instances only a single trypanosome was found in a smear covering the greater part of a slide.

Inoculation experiments showed that 37.5 per cent. of the animals were infected with trypanosomes. Both *T. vivax* and *T. nanum* were found in game, but no conclusions could be drawn as to the presence or absence of these parasites in animals in which they were not found microscopically, since the animals used for the inoculation experiments were not susceptible to either of them.

Different species of animals appear to differ widely in their susceptibility. Trypanosomes were never found in zebra, wildebeest, or bushpig. Waterbuck, bushbuck and kudu were found to be the most heavily infected species, the percentage in these varying from 66.6 to 57.1.

These differences may possibly be referable to some extent to differences of habit of the species, the most seriously infected species being found in thick cover where they would be more exposed to the bites of flies.

A table is given showing the species of trypanosome found in each of the infected animals. In compiling the table the information obtained from the results of inoculation, where that was resorted to, has been utilised. In this way the differentiation between *T. pecorum* and *T. nanum* can be established, these parasites being indistinguishable morphologically. Double infections were not uncommon.

As all the animals shot appeared to be in perfect condition and presented no symptoms of disease, it is probable that their tolerance of the trypanosomes is great, and especially as they have increased in numbers since rinderpest swept through the country.

Examination of domestic stock.—Cattle were found in one village only, and there were only three animals. Two of them appeared to be in good condition, but the third was obviously diseased. These animals represented the remnant of a large herd that had existed there some four or five years before.

The cow in which trypanosomes were found was bred on the Government Farm and had never been outside the limits of the township. Tsetse flies had never been seen within some miles of the place, but *Stomoxys* was abundant in the kraals, and, at certain seasons, various species of *Tabanidae* were common.

A few goats were seen, but these were all dead a few months after.

Glossina morsitans was found around all these villages.

Four goats were under observation for some length of time, but trypanosomes were found in the peripheral blood on rare occasions only.

A dog was found infected with *T. rhodesiense*. This animal came from just over the Nyasaland border. The natives said that it had not been out of the village for over a year, and, as the disease in this species runs a very rapid course, there is no doubt that it was infected locally.

The authors give a brief description of the morphology, pathogenicity, and transmission of the trypanosomes found from which the following details are taken.

T. rhodesiense was found in 16 per cent. of the animals from which inoculations were made.

T. vivax was found in eight waterbuck, one kudu, and three goats. Monkeys, rabbits and rats were found to be insusceptible. The parasite is probably transmitted by *G. morsitans*.

T. nanum.—This trypanosome was found in one bushbuck, three waterbuck, and two goats. It was possibly present in other animals but, in the absence of inoculations, this point could not be settled, as the parasite is not distinguishable from *T. pecorum* morphologically.

The trypanosome is sluggish in its movements and there is not much translation. Inoculation failed in three monkeys, one rabbit, and three rats. There is some evidence to show that it is transmitted by *G. morsitans*.

T. pecorum was found in one bushbuck, one mpala, four waterbuck, two kudu, three dogs, and one wild rat.

Presumably all the animals inoculated—nine monkeys, one rabbit, one guinea-pig, eleven rats, and two mice—became infected.

Evidence was obtained in one case that *G. morsitans* is capable of transmitting this trypanosome, but there is a large amount of evidence that other carriers, and probably more important ones, exist.

Evidence was obtained in one case that possibly Tabanidae are capable of transmitting the infection.

In support of the view that flies belonging to this genus are capable of transmitting trypanosomes it may be mentioned that within the past four years there have been three outbreaks of trypanosomiasis among cattle in districts where tsetse flies may be absolutely excluded. *Stomoxys* and *Lyperosia* are constantly present, and during the rains various species of *Tabanus*, *Pangonia*, and *Haematopota* are common.

A trypanosome which has not yet been identified was found in one bushbuck.

In fresh preparations the parasite was markedly polymorphic. In stained preparations the trypanosome generally resembled the *brucei* or *gambiense*, except for the presence of occasional *pecorum*-like forms. The maximum length was 33·25 microns, the minimum 10·75, and the average 20. Monkeys, rabbits and rats were infected experimentally, but a guinea-pig failed to become infected.

A second trypanosome, the species of which was not known, was found in a dog obtained from the hills on the Nyasaland border.

The parasite in moist preparations appeared as a broad stumpy organism which had no degree of translatory power.

In stained preparations the following details could be made out.

The trypanosome was very broad in comparison to its length, the ratio of breadth to length being 1:4·8. The nucleus was situated at the middle of the body or posterior to it. The blepharoplast was very large and rounded, and situated near the posterior end. Frequently it lay at one edge of the parasite and projected as a small excrescence. The undulating membrane was poorly developed or absent. The cytoplasm contained coarse granules and vacuoles. The average length was 15·8 microns, with a maximum of 20 and a minimum of 10.

A rat showed no trypanosomes up to the thirteenth day after inoculation from the dog, and then it was accidentally killed. The dog died some days previously. The strain was thus lost.

The authors suggest that possibly the trypanosome was the same as that described by MONTGOMERY and KINGHORN in 1909, for which LAVERAN suggested the name *T. montgomeryi*.

Conclusions.

"1. Trypanosomes are of frequent occurrence in game and domestic stock in the Luangwa Valley. At least 37·5 per cent. of the buck harbour parasites.

"2. Six species of trypanosomes were found, viz.: *Tryp. rhodesiense*, *Tryp. vivax*, *Tryp. nanum*, *Tryp. pecorum*, and two others, of which one was possibly *Tryp. montgomeryi*.

"3. *Glossina morsitans*, in nature, transmits two of these trypanosomes, viz.: *Tryp. rhodesiense* and *Tryp. pecorum*, and probably also transmits at least two others, namely, *Tryp. vivax* and *Tryp. nanum*.

"4. Circumstantial evidence exists to shew that *Tryp. pecorum* may be transmitted by biting insects other than tsetse flies."

(125) LAVERAN (A.). Au Sujet du *Trypanosoma pecorum*.—*Bull. Soc. Path. Exot.* 1912. June. Vol. 5. No. 6. pp. 372-375.

The author has already expressed his opinion that the *T. pecorum* is not identical with the *T. congolense* or *T. dimorphon*, basing his view upon the following facts: 1 The serum of a goat which was immune to *T. congolense* and *T. dimorphon* and which was active against these two viruses, was proved to be inactive when mixed with blood containing *T. pecorum*. 2 A goat immune to *T. congolense* and *T. dimorphon* became infected when inoculated with *T. pecorum*.

The author adds a number of fresh details which give additional support to his view. The goat referred to died from the infection, and at the post-mortem the following lesions were found. The goat was only a little more than half the weight that it was three months before death. The inguinal and axillary glands were enlarged. The spleen weighed 90 grammes. No other gross lesions were discoverable. There was severe dry arthritis principally involving the knees, but this lesion ante-dated the inoculation with the trypanosome.

An African sheep which had been successively inoculated with *T. pecuadi*, *T. dimorphon*, and *T. congolense*, and which had acquired immunity to all three was inoculated with blood from a guinea-pig containing the *T. pecorum*. Trypanosomes appeared

in the blood in very small numbers a fortnight later, after which they disappeared from the blood for some time. A dog which was inoculated intraperitoneally with thirty cubic centimetres of blood from the sheep six weeks after trypanosomes had appeared in its blood became infected and died in eleven days. A second dog inoculated in the same manner four months later also became infected and died. A month or two later the sheep began to show signs of ill health and gradually wasted. Death took place nine months after inoculation. The inguinal glands were somewhat enlarged, the peritoneal cavity contained a small quantity of serous liquid, the spleen was enormously enlarged and weighed 820 grammes, and all the other organs appeared normal.

A second goat inoculated with guinea-pig blood containing large numbers of *T. pecorum* became infected on the 10th day. The parasites were present for six days and then disappeared. During the next ten months six dogs were inoculated at intervals and all of them became infected. A seventh dog inoculated two months later failed to become infected.

Two months later the goat was reinoculated with a large dose of guinea-pig blood containing the trypanosome.

A dog inoculated from it three weeks later failed to become infected.

After a further interval of two months the goat was inoculated with blood containing the *T. congolense*, infection resulted and trypanosomes were found in the blood eight days later. Trypanosomes were found in small numbers in the blood subsequently and the temperature varied a little. Up to the time of writing the infection had not proved fatal, but the author states that the reaction may be considered to be exactly like that which would have been obtained with a goat that had not been infected with the other trypanosome.

(126) BOUFFARD (G.). Quelques Considerations d'Ordre Prophylactique concernant le *Trypanosoma cazalboui*. [Prophylactic Measures against *Trypanosoma cazalboui*.]—*Bull. Soc. Path. Exot.* 1912. June. Vol. 5. No. 6. pp. 380-385.

Trypanosoma cazalboui is the most widely spread animal trypanosome in the West African French Colonies. Together with *T. dimorphum* it renders whole tracts of country uninhabitable for equines and bovines. *T. pecaui* is to be found along the banks of all the rivers where they are wooded. Whereas *T. pecaui* is the cause of a sub-acute disease in the horse, is apparently harmless for the donkey, and produces a chronic disease in the ox, *T. cazalboui*, on the other hand, is very virulent for the ox, causing death in as short a period as eight days, but more generally proving fatal in about six weeks.

In the horse it is responsible for a disease which develops very slowly and may last for two to four years. All the Soudanese species are very susceptible.

The disease is rarely encountered in the sheep, but the species is very susceptible to inoculation. The wool-breed in the neighbourhood of Timbuctoo is particularly easy to infect, trypanosomes being very numerous in the blood by the eighth day.

Diagnosis is not associated with difficulty because trypanosomes are always to be found in the blood in eight days, and the morphological characters, and in particular the extreme activity, are sufficiently characteristic. Goats are less susceptible than the sheep, but they are easily infected and are considered by the author to be the "touchstone" in the diagnosis of Souma.

Probably the *G. palpalis* and *G. tachinoides* are the principal agents in the transmission of the disease, but the author has shown that a *Stomoxys*, although not capable of transmitting the disease in the cyclical manner, is by no means a negligible factor. Probably this fly is responsible for the formation of endemic zones outside tsetse areas. These endemic zones are so to speak of an accidental nature and depend upon the introduction of a diseased animal into a stable, the flies acting as carriers to neighbouring animals. These zones may remain infected for long periods provided fresh animals are being constantly introduced into the stables.

The possibility of the existence of persistent centres of Souma should result in a careful examination of all animals moving from place to place. The routes traversed should be provided at intervals with rest houses sufficiently removed from the local animals to obviate the possibility of direct transmission.

The danger is no less great for horses, and the author has seen losses in areas free from tsetse among stallions imported from France.

On several occasions the author has used the sheep as a test animal for inoculation with blood from horses in which the trypanosome could not be found by direct examination, and with considerable success.

The disease is more rapidly fatal in European horses than in the native races, but even in these it lasts ten months, and the trypanosome cannot always be found in the blood.

According to the author the only method of dealing with the disease satisfactorily in breeding stables is to slaughter every animal that is proved to be infected by the inoculation of its blood into a sheep.

The author has made experiments with atoxyl, the benzidine dyes, orpiment, and arsenophenylglycin but has not had a single encouraging result. In every case in which there has been a disappearance of the trypanosome from the circulation there has always been a relapse.

Every horse coming from a tsetse district should be isolated until its blood has been tested experimentally on a sheep. The absence of tsetse is often wrongly interpreted. It by no means indicates an absence of trypanosomiasis, and it may produce a false sense of security.

- (127) BOUET (G.) & ROUBAUD (E.). *Expériences de Transmission des Trypanosomiasés animales de l'Afrique Occidentale française par les Stomoxes*. [Experimental Transmission of Animal Trypanosomes in French West Africa by Stomoxys.]—*Bull. Soc. Path. Exot.* 1912. July. Vol. 5. No. 7. pp. 544-550.

a. *Trypanosoma cazalbouri*.

Experiment 1. A healthy kid was kept beside an infected goat in the open from the 15th September till the 1st October. The ears and the flanks of both animals were clipped in order to facilitate biting of the flies. The kid did not become infected.

Experiment 2. Sixty Stomoxys (*S. calcitrans*, *bouvieri*, and *boueti*) kept in two cages were allowed to bite alternatively and without interval from 28th August to 1st September one infected sheep and one healthy sheep. Trypanosomes appeared in the blood of the sheep on the 2nd September.

b. *Trypanosoma pecauii*.

Experiment 3. A healthy young kid was kept at liberty with an infected kid from 14th September to 1st October. The ears and flanks of both animals were clipped. Flies constantly tormented them. The healthy kid did not become infected.

Experiment 4. Two healthy puppies were kept at liberty with an infected dog and close to some infected cats from 15th September till 29th October. Neither of the dogs became infected in spite of repeated bites.

Experiment 5. A healthy cat was kept at liberty with an infected cat from the 16th to the 22nd October. The cat did not become infected.

Experiment 6. A healthy dog kept with an infected dog for 10 days and after an interval of a fortnight for a further month with another infected dog failed to become infected.

Experiment 7. Five Stomoxys (*calcitrans*) were fed alternately and without interval on an infected guinea-pig and a healthy pig. After the last feed on the infected guinea-pig they were transferred with an interval of five minutes to a healthy guinea-pig. This experiment was repeated daily for six days but neither the pig nor the guinea-pig became infected.

Experiment 8. One hundred Stomoxys (*S. calcitrans*, *soudanense*, *bouvieri*, *glauca*) kept in two cages were fed alternately on a healthy and an infected cat every day for six days. Trypanosomes appeared in the blood of the previously healthy cat the day after the last feeding.

c. *Trypanosoma dimorphon*.

Experiment 9. A healthy kid was kept with an infected goat for four and a half months in the open and was constantly bitten by Stomoxys. The kid failed to become infected.

Experiment 10. One hundred Stomoxys (*calcitrans*) in cages were fed alternately on an infected goat and a healthy goat for five days. The healthy goat did not become infected.

These experiments show that the transmission of these trypanosomes by Stomoxys is nothing like so easy under natural circumstances as would appear from the results obtained when cages are used. Even when cages are used there is a certain amount of variation with regard to transmission of the trypanosomes.

Comparing these results with the following experiments carried out with the Sahara virus the role of the Stomoxys in the transmission of virus naturally transmitted by Glossina is obviously a secondary one.

Transmission of the Saharan Viruses by Stomoxys.

a. Trypanosoma soudanense.

The virus used was of the Tahaga type and was obtained from the valley of the Niger.

1. Transmission by immediately successive bites.

Experiment I. A hundred flies (*S. calcitrans*, var. *soudanense*, *S. bouvieri*, and *S. vittata*) were fed alternately without interval on an infected dog and a healthy rat for two days. The rat became infected a fortnight later.

Experiment II. Similar to Experiment I. with corresponding result.

Experiment III. Fifty flies fed alternately on an infected dog and a healthy guinea-pig with an interval of half an hour. The guinea-pig died a week later without showing trypanosomes.

Experiment IV. Repetition of Experiment III. using an infected rat in the place of the dog. The guinea-pig was not infected a month later.

Experiment V. Flies fed alternately without interval on an infected rat and a healthy dog for three days. The dog became infected a week later.

Experiment VI. Repetition of Experiment V using an infected dog in the place of the rat. The healthy dog was found to be infected a week later.

2. Natural mechanical transmission.

Experiment VII. A young healthy dog was freed from fleas, washed with cresyl, and kept during the day in the open with an infected dog. During the night the animal was placed in a fly-proof cage. The dogs were kept in contact for eight days. On the sixth day after the separation of the two animals the healthy dog showed trypanosomes.

3. Transmission after an interval of at least 24 hours.

Experiment VIII. About eighty of the flies used in the previous experiment were allowed to bite a healthy dog for six days commencing one day after they had had their last feed on the infected dog used in experiment VII. The dog was found to be infected a month later.

The dog was examined up to the end of the following month, but trypanosomes were not discoverable microscopically in its blood until two days before its death. A month before death occurred the serum of this animal if left standing at the temperature of the laboratory became a rich culture of the trypanosomes within a few hours.

A second experiment of this kind in which the infected flies were kept for forty-eight hours before being placed on the healthy animal was abortive owing to the escape of the dog.

b. *Trypanosoma evansi*.

In a series of twelve experiments exactly analogous results were obtained with this trypanosome.

- (128) BRIEGER (L.) & KRAUSE (M.). *Chemotherapie bei Trypanosomeninfektion (Trypanosoma brucei) nach Verabreichung per os.* [The Treatment of Nagana by means of Drugs administered by the Mouth.]—*Berlin Klin. Wochenschr.* 1912. July 29. Vol. 49. No. 31. pp. 1453-1455.

In a previous paper the authors have shown that bodies of the safranin group possess a specific trypanocidal action, and KRAUSE working in conjunction with WEBER has adduced evidence that this action depends upon the side chains in the molecule, the removal from or the addition to the side chains of a group causing the drug to become increasingly or decreasingly active.

The authors state that the failure of the drugs employed by other authors to effect a radical cure is probably due to the fact that they act on the parasites present in the blood only and are too rapidly split up, the trypanosomes in the organs and tissues escaping their action. Their object therefore was to discover some substances which were stable and which would be able to reach all tissues. They claim to have found such substances belonging to the Safranin and Eurhodin groups.

The characteristic features of these dyes are that they contain in their molecules a combination of trivalent and pentavalent nitrogen and that they possess an extraordinarily active trypanocidal action, even when administered by the mouth. They are quite stable and in the pure state non-toxic. Thirty or more grammes may be given to the human subject in doses of $\frac{1}{4}$ to 1 gramme daily without any secondary effects. The substances contain no arsenic.

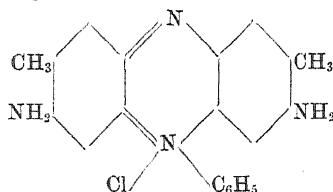
The animals used for the experiments were rats and guinea-pigs and four strains of *T. brucei* were employed. The results were the same with each of the strains. Series of infected animals were fed with the drugs, some being kept as controls in every case. All the animals receiving the substances were cured, that is to say they remained free from trypanosomes for months. The drugs were mixed with moistened bread for the rats and with moist bran for the guinea-pigs, and the dose was from 0.05 to 0.1 gramme. The feeding was continued for 4-6 weeks and every other day food was given with the admixture of the dyes.

Rats which had been free from trypanosomes for months were more difficult to infect than fresh animals, and the infection in such rats was more easily overcome. It was found that all compounds were not equally valuable. Some compounds which were actively trypanocidal entirely lost that power when their molecule was a little altered. With the most suitable compounds the

authors were able to free guinea-pigs from trypanosomes six days after inoculation and twenty-four hours before death, and to produce an absolute cure with further doses. The control guinea-pigs in the same experiments died in about seventeen days.

The sera of uninfected animals which were given the drug daily for some time exercised no protective or curative effect.

A substance having the following composition was very active :



Apparently the symmetrical arrangement of the methyl and the amido groups is connected in some way with the action. If one of the methyl groups be removed not only is the colour of the substance changed, but the trypanocidal action is nearly destroyed.

If two methyl groups are introduced into one of the amido groups serious sequelae follow the administration of the compound. The removal of both methyl groups restores the trypanocidal action, but the substance so obtained is not so active as the first compound mentioned.

The effect of methyl and amido groups on the trypanocidal action of drugs has already been shown by Krause in the dyes of the triphenylmethane group.

A number of other details are given by the authors regarding the effects produced by modifying the side chains of the molecule. In some cases there is loss of trypanocidal effect, in others toxic effect are produced, and so on.

Other bodies of the safranine group, such as indulin-scharlach, were tried, but their trypanocidal action was weak, as also was the action of the eurhodin group.

- (129) CASTELLI (G.). *Chemotherapeutische Versuche über die Wirkung des Kakodyl und Arrhenal bei experimentellen Spirillen- und Trypanosomenerkrankungen.* [Experiments with Kakodyl and Arrhenal in Experimental Spirillosis and Trypanosomiasis.]—*Arch. f. Schiffs- und Trop. Hyg.* 1912. Sept. Vol. 16. No. 18. pp. 605-619.

The author's experiments had for their object the determination of the toxicity of these two compounds for the mouse, fowl and rabbit, and their curative effect on recurrent fever, trypanosomiasis (nagana), spirochaetosis of the fowl, experimental syphilis and yaws.

It was found that the maximum safe dose (dosis tolerata) of sodium kakodylate for the mouse by subcutaneous injection was 0.02 g. per 20 g. body weight. For the fowl intramuscularly the dose was 0.6 per kilo, and for the rabbit by intravenous injection 0.3 g. per kilo.

The following results were arrived at in experiments with arrhenal. Maximum safe dose for the mouse subcutaneously

0.5 g. per 20 g. body weight; 1.6 g. per kilo. for the fowl intramuscularly; and 0.5 g. intravenously for the rabbit.

Experiments to test the curative value of the compounds in the diseases mentioned showed that they were absolutely without effect.

- (130) LAVERAN (A.) & ROUDSKY (D.). *Au Sujet de l'Action de l'Akridine (Diphénylméthane) sur Trypanosoma lewisi et Trypanosoma duttoni.* [The Action of Acridine on *T. lewisi* and *T. duttoni*.]—*Compt. Rend. Soc. Biol.* 1912. July 26. Vol. 73. No. 27. pp. 172-175.

In a previous publication the authors have shown that oxazine and acridine have a remarkable affinity for the substance of the centrosome of trypanosomes and they have come to the conclusion that the destruction of the centrosome is in all probability due to a process of auto-oxidation.

According to KUDICKE the centrosome of *T. lewisi* in rats treated by acridine disappears in a definite fashion. According to this author one portion of the centrosome becomes displaced towards the anterior portion of the body while the origin of the flagellum undergoes no alteration. The portion which alters its position is re-absorbed or eliminated *in toto*. KUDICKE has never observed the penetration into or the fusion of this portion of the centrosome with the nucleus. If acridine is administered during the acute stage when multiplication forms of the parasite are numerous the process according to KUDICKE is different. At the moment when certain trypanosomes are undergoing division the centrosome itself does not divide, one of the daughter trypanosomes thus being left without a centrosome. This abnormal division which has already been described by WERBITZKI would be the starting point of the acentrosomic varieties of the trypanosome.

Up to the present the authors have not observed the changes described by KUDICKE, the explanation being that they have only studied the action of oxazine on pathogenic trypanosomes, and the peculiar phenomenon observed by KUDICKE apparently occurs only in animals infected with non-pathogenic trypanosomes and treated by acridine. A more recent series of experiments carried out with acridine on rats infected with *T. duttoni* has confirmed KUDICKE's results. A rat inoculated with *T. duttoni* showed numerous trypanosomes in its blood on the fourth day after inoculation and it was given a quarter of a milligramme of acridine in a freshly prepared solution. On the following day a certain number of the trypanosomes were observed to possess no centrosome while in others the centrosome was free in the cytoplasm. The position of the free centrosome was variable. The posterior extremity of the flagellum was not altered in position but in a number of cases showed a slight thickening. Two days later the rat received a second injection of acridine, the same dose being administered. On the following day the number of acentrosomic trypanosomes was increased and the rat was killed for the purpose of inoculating a fresh one. It appears to be probable that under the action of acridine the centrosome becomes more friable and as a result of the movements of the flagellum becomes detached and is thus left

free in the cytoplasm. The authors have examined trypanosomes with free centrosomes coloured by oxazine or acridine *in vitro*, and they have been able to determine that the movements of the free centrosomes are purely passive. These centrosomes are gradually absorbed thus producing acentrosomic trypanosomes. Before disappearance is complete the centrosome is in some cases reduced to a collection of fine granules which can be stained *in vitro* by acridine or oxazine. The absence of such effects of acridine upon the centrosomes of pathogenic trypanosomes may be explained as follows:—These centrosomes which are much smaller than those of *T. lewisi* and *T. duttoni* offer less resistance to the traction exerted upon them by the flagellum and consequently are less likely to become detached. Acridine also has far less effect upon the centrosomes of the pathogenic trypanosomes than upon those of *T. lewisi* and *T. duttoni*. On the other hand oxazine has far less action on the latter than upon the majority of the pathogenic trypanosomes.

- (131) PECAUD (G.). Contribution au Traitement des Trypanosomaises Animales. [The Treatment of Animal Trypanosomiasis.]—*Bull. Soc. Path. Exot.* 1912. June. Vol. 5. No. 6. pp. 385-389.

Cattle appear to be more susceptible to orpiment than horses, and the maximum dose should not exceed 7-8 grammes per 100 kilo. live weight. The administration of the drug is difficult in cattle. If given in the form of a ball there is a risk of the ball resting in the rumen or abomasum and setting up severe inflammatory conditions which may lead to gastric fistula and necrosis of the stomach and skin. For this reason the author administered the orpiment in the form of an electuary.

In dealing with cattle the following plan was resorted to: The electuary was given every two days, the dose to commence with being about 4 grammes per 100 kilog. The dose was gradually increased until the maximum dose was given at the fourth or fifth administration. This last dose was repeated once.

As a general rule the trypanosomes disappeared from the blood after the first dose, and this was particularly the case with *T. caalbouvi*.

In some instances they reappeared eight to ten days after the first series of doses, in which case the treatment was repeated.

The mixed method of treatment involves the alternate administration of orpiment and atoxyl, the latter being given subcutaneously in doses of 2 to 3 g. at a time.

The author prefers to use orpiment alone as it is just as effective and less costly, but orpiment may be replaced by an injection of atoxyl when the animal shows symptoms of poisoning.

In any case no definite rules can be laid down, cases must be treated on their merits. Great care has to be taken in administering orpiment because there is very little difference between the toxic and the therapeutic dose, and it is of the utmost importance to use the pure precipitated drug.

Relapses are not uncommon, and in such cases treatment has to be commenced over again.

The author has obtained similar results with tartar emetic, and he finds that it is easier to administer. He follows the plan already advised by BRODEN and RHODAIN, and administers the drug subcutaneously, using a 2 per cent. solution for the purpose. In the majority of cases four or five administrations of 1 to 2 g. suffice.

It would appear that orpiment is more effective against *T. cazalboui*, while emetic is more effective with *T. dimorphon*.

Neither drug appears to have any permanent effect on *T. pecaudi* in the horse.

The following are the results obtained and in every case the animals have been followed for a period of two years or more.

Cattle.—*T. dimorphon*.—Emetic. Eight cases treated, six recoveries and two deaths.

Orpiment alone.—Twenty-four cases treated, eighteen recoveries, three deaths, and three cases of gastric fistula as a result of administering the drug *in bolus*.

Orpiment-atoxyl.—Seven cases treated, one recovery, four deaths, and two deaths from gastric fistula.

Horses.—*T. pecaudi*.—Atoxyl-orpiment.—Three cases treated, all failures.

T. cazalboui.—Five cases treated, one death, two successes, one relapse successfully treated, and one relapse which was not treated and proved fatal.

Donkeys.—*T. cazalboui*.—Two cases treated. One death, and one success, but the animal aborted.

Comparing the mortality among treated and untreated animals the author finds that while among untreated animals the mortality has been 90 per cent. and none of the pregnant females have had a normal parturition, in treated animals (excluding accidents) the mortality has been 26 per cent., and abortion has occurred in 37.5 per cent. of cases.

The author adds that in every case the animals put under treatment had been affected for a short time only.

- (132) MATTES (W.). Agglutinationserscheinungen bei den Trypanosomen der Schlafkrankheit, Nagana, Dourine, Beschalseuche und des Kongokustenfiebers, unter Berücksichtigung der Farbemethoden, der Morphologischen und biologischen Verhältnisse der Erreger. [Agglutination of the Trypanosomes of Sleeping Sickness, Nagana, Dourine, and Congo Coast Fever, and Consideration of the Staining Characters and Morphological and Biological Characters of the Organisms.]—*Centraltb. f. Bakt.* 1. Abt., Orig. 1912. Aug. 10. Vol. 65. Nos. 6-7. pp. 538-573.

The procedures adopted and the results obtained by a number of other observers are briefly referred to and a somewhat extensive description of the morphology, developmental forms, pathogenicity, vitality, cultivation, symptoms, lesions, and staining characters of the various species of trypanosomes used are given before the author proceeds to the description of his own experiments in connection with the agglutination of trypanosomes.

The following species of trypanosomes were used in the experiments: *T. gambiense*, *T. brucei*, *T. equiperdum*, *T. congolense*,

and the trypanosome of dourine. According to the author *T. equiperdum* is the cause of dourine (*Beschläseuche*) which occurs extensively in Europe, and the parasite referred to as the trypanosome of dourine is the organism responsible for the disease in Algeria.

The author points out the differences between the agglutination test as carried out with bacteria and with trypanosomes, emphasising the fact that trypanosomes for the purpose must be obtained from an infected animal and also the supreme importance of absolute sterility of all the instruments and vessels.

In the majority of cases the author obtained his suspension of trypanosomes by bleeding animals at the height of infection into citrate solution. He found it inadvisable to chloroform the animal, because this lessened the amount of blood obtainable, and he also feared that the chloroform might have some effect upon the agglutination. The mixture thus obtained was centrifuged in the water centrifuge because it was observed that if the electric centrifuge were used at 4,000 revolutions per minute the trypanosomes were so clumped together as to be useless. Following LANGR'S procedure the author found it difficult to obtain a suspension of trypanosomes free from red blood corpuscles, he therefore devised the following methods. The tubes used for centrifuging the diluted blood were about 5 mm. in diameter, and the lower end was drawn out into a capillary tube about 10 cm. long and sealed up. During centrifugalisation the tubes were carefully packed round with wool to prevent breakage. After complete sedimentation of the corpuscles the capillary tube was broken at the junction of the sedimented corpuscles and the layer of liquid containing the trypanosomes, the upper end of the tube being closed to prevent the liquid from flowing out. The trypanosomes thus obtained were thrice washed.

The second plan devised by the author was to stop the centrifuging at the moment when the great majority of the red cells had been thrown down, but the trypanosomes were still to a great extent in suspension. The supernatant liquid containing the trypanosomes and a few red corpuscles was drawn off and again centrifuged, the process being continued till sedimentation was complete. A delicate capillary pipette was then introduced and the trypanosomes stirred up into the liquid by careful rotation of the point of the pipette around the tube just above the layer of trypanosomes. Proceeding in this way, and if necessary adding more salt solution and re-centrifuging, the majority of the trypanosomes could be obtained.

The suspension so obtained had then added to it five to ten per cent. of 15 per cent. formalin in salt solution. Experiments were made without success to replace the formalin by carbolic acid, cresol, chloroform, and absolute alcohol.

About two to three cubic centimetres of suspension are to be obtained from a large mouse.

Spontaneous agglutination of trypanosomes in the suspension is likely to occur if too large a proportion of formalin be added, and also if the suspension be allowed to stand before the formalin is added. The sera used in the tests were obtained from man, the

horse, ox, sheep, goat, pig, rabbit, guinea-pig, and mouse, and in no case was any preservative added.

The mixtures of serum and suspension were kept sterile by means of plugs during the test, and the tubes were incubated for twelve or more hours at 37° C. In some cases it was necessary to incubate for thirty to forty hours before any reaction became evident. The author found that normal sera were capable of agglutinating trypanosomes in much higher dilutions than normal sera generally agglutinate bacteria. Tests were made with each of the species of trypanosome using normal sera from the animals mentioned, but in no case was any agglutination observed in dilutions higher than 1 in 100. The sera of the horse and the ox were found to possess the highest power, and in practically no other case was any agglutination observed in dilutions higher than 1 in 10.

Agglutinating power of immune sera. In order to obtain as large an amount of agglutinin as possible the author first inoculated his animals with either dead trypanosomes, or trypanosomes that had lost a great deal of their vitality through the blood being allowed to stand after withdrawal for a time, and then again inoculating with living fully virulent trypanosomes after an interval of about a week. In the case of the least pathogenic trypanosome—the *congolense*—a third inoculation was given. Blood was withdrawn at intervals and the agglutinating power tested, and the animals were killed when seriously ill. It was found that there was a somewhat rapid elevation in the amount of agglutinin present.

Tests were made at intervals with homologous and heterologous trypanosomes. From these it was found that the reactions are not specific, a nagana-immune serum agglutinates not only *T. brucei* but all the other species used in the tests, but the agglutination with the homologous strain occurs in higher dilutions than with the heterologous strains. For example, the serum of a rabbit inoculated with nagana agglutinated the *T. brucei* in a dilution of 1 in 3,000, the dourine trypanosomes in dilutions of 1 in 2,000, and the *T. congolense* and *T. gambiense* in dilutions of 1 in 1,500.

The author found that agglutinin is present in exudates in the serous cavities as well as in the blood.

The sera of two horses that had recovered from dourine were tested with suspensions of the different trypanosomes and similar results were obtained.

The results were not influenced by inactivating the serum by heating for some hours at 60° C., nor did heating the suspension have any effect on the results. Exactly similar results were also obtained when the suspension was cooled to — 25° C. for three hours.

A number of experiments were made in which the serum was diluted and the suspension made with salt solution that had been rendered radio-active. In two instances there was spontaneous agglutination in suspensions so prepared without the addition of any serum. The experiments made with sera lead the author to conclude that the agglutination occurred in much higher dilutions and more rapidly when radio-active salt solution was used than when ordinary salt solution was employed.

- (133) SCHILLING (C.). Ein neues Immunisierungsverfahren gegen Trypanosomen-Infektionen. [A New Method of Immunising against Trypanosome Diseases.]—*Deut. Med. Wochenschr.* 1912. Aug. 22. Vol. 38. No. 34. pp. 1579-1580.

The principle upon which the method is based is the inoculation of susceptible animals with trypanosomes which have been killed by chemical substances, but which, according to the author, still retain their power of provoking the formation of antibodies.

Experiments have been made with two horses using the *T. brucei*. It would appear that the trypanosomes are obtained free from blood and treated for two hours with an equal quantity of 1 : 400 solution of tartar emetic. This liquid is centrifuged and the sediment used as the antigen.

The first horse treated received seven injections, some subcutaneous and some intraperitoneal, of trypanosomes treated in this way obtained from a total of fifty infected rats, the injections being spread over a period of ten weeks. A week after the last injection the horse received an intravenous inoculation of 0.5 cc. of mouse blood containing *T. brucei*. The horse failed to become infected.

The second horse was treated in a similar manner, but only five injections of dead trypanosomes were given. When tested with 0.5 cc. of mouse blood containing trypanosomes the animal failed to become infected.

No definite results have been obtained in experiments in which increasing quantities of serum from treated animals were mixed with trypanosomes and inoculated into the peritoneal cavity of mice, with the object of standardising the serum.

The duration of the immunity was tested by reinoculating a number of mice at intervals after they had been protected and inoculated once. The results were inconstant. Whereas two mice were reinfected after an interval of two months, two others resisted this inoculation and two further inoculations at intervals of one and two months.

- (134) DUKE (H. L.). Antelope as a Reservoir for *Trypanosoma gambiense*.—*Proc. Roy. Soc.* 1912. July 25. Series B. Vol. 85. No. B. 579. pp. 299-311.

In considering the problem of the prolonged infectivity of *G. palpalis* on the shore of Lake Victoria Nyanza the subject must be approached from three points of view, (1) some idea of the percentage of infected wild flies must be obtained from time to time in order to gauge the efficiency of the reservoir, (2) the wild animals frequenting the Lake shore must be examined for *T. gambiense*, and (3) the effect of *T. gambiense* on these animals should be studied in the laboratory.

It has been pointed out in the Report covering the period March, 1911, to January, 1912, that the percentage of infected flies along the Chagwe coast line is 0.014 per cent. During the interim no natives have been allowed in the neighbourhood of the fly except those in the employ of the laboratory.

On Damba Island in May, 1911, CARPENTER obtained infection of a monkey with *T. gambiense* using 880 wild fly, and two out

of four animals shot on this island were found to harbour the trypanosome. The conditions on this island are exceptional in that the fly and antelope are frequently brought into contact.

Under such conditions it would appear necessary for the antelope to remain infected over a very long period in order to keep up the infectivity of the fly, and nothing short of a very rapidly acquired immunity to the disease on the part of the antelope could prevent a relatively large number of fly being infective at any given time. On the main land the opportunities for the fly to bite these animals are much less frequent, and, bearing in mind the small percentage of flies capable of carrying the trypanosome, it becomes of great importance to know how long an antelope can remain infective.

It has been shown that these animals may remain infective to fly for a period of at least 10 months. Up to May, 1911, the evidence of infectivity afforded by the presence of trypanosomes in the laboratory bred *G. palpalis* fed upon the buck proved sufficiently regular without resorting to blood injection. The experiments given in this paper represent the continuation of these investigations up to January, 1912. During this period laboratory bred *G. palpalis* were fed upon the antelope and their blood was also tested by injection into susceptible animals. The injection test gradually came to supersede the employment of clean flies.

Details are given of fly experiments and injection experiments with six antelope, and the longest period for which one of these was found to remain infective was 22 months after its original infection with laboratory bred *G. palpalis*.

Brief details are also given regarding experiments which were undertaken to ascertain whether the serum of these antelope had acquired any immunising properties. The author concludes that there is some evidence to show that an antelope which has ceased to be infective for *T. gambiense* acquires some degree of immunity against re-infection.

(135) KINGHORN (Allan) & YORKE (Warrington). On the Transmission of Human Trypanosomes by *Glossina morsitans*, Westw.; and on the Occurrence of Human Trypanosomes in Game.—*Ann. Trop. Med. & Parasit.* 1912. March 29. Vol. 6. No. 1. A. pp. 1-23.

The authors' summary is as follows:—

" 1. The human trypanosome, in the Luangwa Valley, is transmitted by *Glossina morsitans*, Westw.

" 2. Approximately 5 per cent. (4.76) of the flies may become permanently infected, and capable of transmitting the virus.

" 3. The period which elapses between the infecting feed of the flies and the date on which they become infective, is approximately fourteen days.

" 4. An infected fly retains the power of transmitting the disease during its life, and is infective at each meal.

" 5. Mechanical transmission does not occur if a period of twenty-four hours has elapsed since the infecting meal.

" 6. Some evidence exists to shew that in the interval between the infecting feed and the date on which transmission is possible, the parasites found in the flies are non-effective.

"7. *Glossina morsitans*, in nature, has been found to transmit the human trypanosome.

"8. Certain species of buck, viz., waterbuck, hartebeest, mpala, and warthog, have been found to be infected with the human trypanosome.

"9. A native dog has been found to be infected with the human trypanosome."

- (136) DUKE (H. L.). Observations on Fowls and Ducks in Uganda with Relation to *Trypanosoma gallinarum* and *T. gambiense*. (With a Note by Miss Muriel ROBERTSON.)—*Proc. Roy. Soc.* 1912. August 20. Series B. Vol. 85. No. B. 580. pp. 378-384.

In the course of a considerable number of experiments carried out to test the duration of infectivity of antelopes for *T. gambiense* Uganda fowls were employed to nourish the flies during the earlier days of the experiment. In the course of routine dissections of laboratory bred flies from these experiments a curious crithidial type of flagellate was noticed in the hinder part of the mid-gut. In morphology and movement the organisms were immediately distinguishable from the developmental stages of the *T. gambiense* in *G. palpalis*. As some of the laboratory antelope had been found to be infected with *T. ingens* it appeared possible that the flagellates might represent developmental stages of this trypanosome. On the other hand suspicion fell upon *T. gallinarum*, although microscopic examination of the fowls used had always proved negative. The connection between *T. gallinarum* and these crithidia was established by finding them in clean laboratory bred flies fed upon the suspected cocks and by the subsequent discovery of the trypanosomes in the cocks' blood. The trypanosomes are very rare in the peripheral circulation.

The following is a summary of Miss ROBERTSON's note on the morphology of the parasites:—In many respects the appearance of *T. gallinarum* in *G. palpalis* is exceedingly characteristic. The trypanosomes are, as a rule, situated in the middle and posterior portions of the mid-gut. The motion of the flagellate in the live state is very striking, the body in the vast majority of cases is quite stiff through about two-thirds of its length and is often broad and relatively massive. The flagellum is much thicker than in mammalian trypanosomes. Progression takes place in straight lines, and in the more slender individuals may occasionally be reversed in direction. The breadth of the body is subject to variation, and as it is impossible to tell when the flies first receive the trypanosomes it is impossible to give the exact sequence of the forms in the cycle. Apparently, however, the massive broad forms gradually give place to smaller and more slender individuals, some of which show considerable lengthening of the body.

True trypanosome phases have not been observed. Although binary fission appears to be the usual method of multiplication, multiple division is not uncommonly observed.

Morphology.—Films were fixed wet with Schaudinn's fluid and stained with iron-haematoxylin and haemalum.

The protoplasm especially in the broader forms is very dense and granular. The tropho-nucleus is of a common trypanosome

type, consisting of a large central karyosome surrounded by a clear space which is bounded in turn by a delicate membrane. Fine rays pass from the karyosome to the membrane.

The kineto-nucleus lies close to and generally in front of the tropho-nucleus. It is relatively large and presents the somewhat curious double appearance of two closely-apposed granules lying one behind the other. This does not seem to be in any way a case of precocious division. The blepharoplast proper, *i.e.*, the minute granule actually at the origin of the flagellum is usually obscure but can be seen in favourable specimens.

Another structure not usually observed in trypanosomes is a granule situated towards the posterior end of the cell. The outline of this granule is rarely very sharply defined and in some instances it appears to be attached by a fine line to the nucleus or to the blepharoplast. The granule divides when the parasite divides.

A few experiments were undertaken to ascertain whether ducks were capable of acting as a reservoir for *T. gambiense* and whether newly arrived ducks were capable of infecting laboratory bred flies with flagellates. The results in both cases were negative.

(137) MESNIL (F.) & BLANCHARD (M.). Infection des Poules dues aux *Trypanosoma gambiense* et *Trypanosoma rhodesiense*. [Infection of Fowls with *T. gambiense* & *T. rhodesiense*.]—*Compt. Rend. Soc. Biol.* 1912. June 14. Vol. 72. No. 21. pp. 938-940.

The authors inoculated four fowls each with *T. gambiense* and *T. rhodesiense*. The inoculations were successful in every instance in the former and in three of the birds in the latter. Trypanosomes were never found in moist preparations of blood, but the blood was proved to be infective by inoculation. In two birds a few trypanosomes were seen in the leucocyte layer of blood taken at the time of death and centrifuged. Three of the fowls inoculated with *T. gambiense* died in one to three months. In the fourth bird the disease lasted for a month and rats subsequently inoculated failed to become infected, and the bird was thought to have recovered. It was reinoculated and died ten days later, its blood proving infective for mice.

Two of the birds infected with *T. rhodesiense* died in about two months.

The blood of the third was proved to be infective up to the fifty-fifth day, and the bird died on the eighty-third day, its blood being then non-infective. Death was due to pasteurellosis. The fourth fowl died of pasteurellosis without becoming infected.

There were lesions involving the eyes of the birds that died of the infection in both cases.

The serum of the birds infected with *T. rhodesiense* mixed with the homologous trypanosome retarded the evolution of the disease in a mouse for three or four days, but had no action on the heterologous trypanosome. The serum of birds infected with *T. gambiense* had no effect upon either.

- (138) BAYON (H.). The Cultivation of *Trypanosoma rhodesiense*, Stephens and Fantham.—*Proc. Roy. Soc.* 1912. August 24. Series B. Vol. 85. No. B. 581. pp. 482-483.

The author has been able to cultivate *T. rhodesiense* successfully on the following media, (1) CLEGG's *Amoeba* agar to which is added twice its amount of rabbit's blood which has been frozen and thawed rapidly so as to cause the haemoglobin to diffuse into the serum; and (2) the following formula:—agar 15 grammes, glucose 10 grammes, water 1,000 grammes, and twice the volume of rabbit's blood.

The blood containing the trypanosomes was placed on the agar which forms a diffuent mass at the bottom of the tube. The tubes were incubated at 22° to 25° C. Multiplication could be observed under the microscope, and the cultivated trypanosomes remained actively motile up to the twenty-first day of cultivation. They were virulent up to the eighth day of culture. In certain cases rosette forms were observed and true-brood forms have also been distinguished. In some of the tubes rounded forms appeared on the third to the fifth day; these are being separately studied. The percentage of successes varied from one tube in twenty-five to six in six.

- (139) YORKE (Warrington) & BLACKLOCK (B.). A Note on the Morphology of a Strain of *Trypanosoma equiperdum*.—*Brit. Med. Jl.* 1912. August 31. p. 473. With 1 chart and 13 text-figures.

The occurrence of posterior nuclear forms of this parasite had already been noticed by the authors in smears of guinea-pig blood infected with this trypanosome some months before the present observations were made.

More recently the authors obtained a fresh strain of dourine virus from SCHILLING, the strain having been running in his laboratory for many years.

Morphology. Between the long thin forms with free flagellum and short stumpy forms without free flagellum there is a variety of intermediate forms.

A thousand parasites from a white rat, drawn and measured over a period of eight days before its death, had an average length of 26·7 microns. The longest measured 36 and the shortest 15 microns.

The forms of the parasite in which the nucleus is observed to lie posterior to the centre are the short stumpy types which have little or no free flagellum. They have been observed in rabbits, guinea-pigs and rats—the only animals that have been experimented with. The nucleus varies in position from being just posterior to the centre of the body to close to the blepharoplast. A definite posterior position of the nucleus was found in 3·8 per cent. of a thousand trypanosomes. Many of the parasites have very marked granules.

- (140) WENYON (C. M.). The Insufficiency of the Posterior Nucleus as a Specific Distinction in *Trypanosoma rhodesiense*.—*Jl. Trop. Med. & Hyg.* 1912. July 1. Vol. 15. No. 13. pp. 193.

The figures of *T. rhodesiense* given by STEPHENS and FANTHAM reminded the author of *T. pecaui*, and on looking through some films of rat blood containing *T. pecaui* which he made in the Sudan in 1908 he found that forms with the trophonucleus near to the kintonucleus are fairly common, while a small percentage of the comparatively broad trypanosomes have the nucleus at the posterior end of the body.

In the light of the diagnostic importance of this arrangement of the nucleus in *T. rhodesiense*, as emphasised by STEPHENS and FANTHAM, the author thought it worth while to place on record the present observations.

In the case of *T. pecaui*, the forms with the nucleus near the posterior end occurred both in the blood of inoculated rats and in the blood of the original host—a donkey. In the latter the author has not found a specimen of the trypanosome with the nucleus on the posterior side of the kintonucleus, but thinks it probable that prolonged search would reveal such, since in many the nucleus is right up against the kintonucleus. In the case of *T. rhodesiense* the posterior nuclear forms occur in sub-inoculated rats only, whereas in *T. pecaui* they occur in the donkey also. This is probably dependent upon the infection, which is always very small in man, while it was heavy in the donkey. In the case of both these trypanosomes it is perhaps incorrect to speak of either man or donkey as the true host. In neither case is there any sign of adaptation, for the infection nearly always brings about a fatal termination. The true host is the reservoir from which man and the donkey become infected.

It is quite possible that one and the same trypanosome would show certain morphological variations in the same host, provided that in one district the host had become more adapted to the parasite than in the other. It would be interesting to know whether *T. pecaui* shows posterior forms in the reservoir host, but this has apparently not yet been discovered.

Drawings of nine trypanosomes are given and the author points out that the broad forms with the posterior nucleus divide and give rise to trypanosomes of the ordinary type.

- (141) ROUDSKY (D.). Action Pathogène de *Trypanosoma duttoni* Thiroux, et Lésions Provoquées chez le Rat par ce flagellé. [The Pathogenic Effect of *Trypanosoma duttoni*, and the Lesions caused by the Organism in the Rat].—*Compt. Rend. Soc. Biol.* 1912. July 26. Vol. 73. No. 27. pp. 170-172.

In a previous publication the author has shown that *T. duttoni* is capable of causing death in the rat. This pathogenic effect, which was practically constant in the first passages in series from rat to rat, gradually became attenuated as the number of passages increased. The author has made 53 passages and every rat has become infected, but death has been exceptional and haemoglobinuria very rare. The infection itself has become more brief since

the first passage. The pathogenic power acquired by *T. duttoni* by changing the vertebrate host is modified in a quite different manner to that of *T. lewisi* acclimatised for the mouse, the latter showing a progressive increase of virulence.

During the first passages from rat to rat *T. duttoni* showed characters different from those observed in the mouse. The posterior extremity became elongated to the extent of exceeding in some individuals the length of the anterior part. This posterior extremity was composed either of periplasm alone or of two layers of periplasm separated by a small amount of cytoplasm. In certain instances the elongated posterior extremity resembled a second flagellum in appearance. This morphological character suddenly disappeared at the 9th passage, the parasite losing its pathogenic power at the same time.

The centrosome in practically every parasite appeared bi-lobed.

Lesions were to be found in a number of the viscera of rats which died showing symptoms of haemoglobinuria as a result of infection with *T. duttoni*.

The hepatic cells showed a slight amount of coagulative necrosis around the central veins resulting from pressure by the engorged capillaries. Cellular infiltrations, consisting principally of mononuclear leucocytes, were found around the portal spaces. In one instance the author found giant cells.

In the kidneys a considerable number of the tubes were filled with an amorphous material. In some portions of the tubes there was complete destruction of the epithelium, the lumen being filled with a finely granular material which contained a number of desquamated cells more or less altered in appearance.

The parenchyma of the spleen was infiltrated with a large quantity of blood.

To the naked-eye reddish areas could be observed in the parenchyma of the lungs, the acini in these places being distended with an oedematous fluid.

The author concludes that the lesions produced may be due to a toxin developed by *T. duttoni* to which the rat is susceptible.

- (142) SCHEPILEWSKY (E.). Fadenförmige Anhängsel bei Trypanosomen. [Thread-like Appendages in Trypanosomes.]—*Centralbl. f. Bakt.* 1. Abt. Orig. 1912. July 3. Vol. 65. Nos. 1-3. pp. 79-83. With 1 plate.

After a brief reference to the morphological characters of the *T. brucei* and *T. equiperdum*, the author passes to a consideration of the work of WASIELEWSKI and SENN, who first observed marked elongation of the posterior extremity of trypanosomes in rat blood, and who considered the elongation to be an artefact due to the spreading of the blood. WENDELSTADT and FELLNER who also observed this elongation did not agree that it should be referred to an artefact because it was not observed to be all in one direction. The work of v. PROWAZEK, DOFLEIN, LEPORSKY, YAMAMOTO and others, is mentioned.

The author's own observations have been made with dark-ground illumination, and the two trypanosomes used for the

observations were those mentioned above, the strains having been kept in guinea-pigs and mice. The blood used for examination was mixed with salt solution in such proportions that under the microscope there was a certain amount of space between the blood corpuscles. The preparations had to be examined immediately they were made as the trypanosomes died in about half an hour.

With dark ground illumination it can be quite clearly made out that the bodies of the parasites are surrounded by a thin refractile envelope which is sharply defined both externally and where it abuts on the cytoplasm. This envelope (pellicle) extends to the anterior end of the flagellum giving it a double-contoured appearance. The double contour seems to disappear at the extreme end of the flagellum under a comparatively low magnification, but it can be easily made out under the oil immersion. For this reason it cannot be asserted that the two trypanosomes under consideration have in reality any free flagellum.

No structure can be made out in the endoplasm with dark-ground illumination while the trypanosome remains alive, and the nucleus is not visible. Sometimes one or more refractile particles can be made out in the cytoplasm. Immediately the trypanosomes die the whole of the protoplasm becomes granular, and the nucleus appears as a dark spot.

The author has been able to observe details which do not appear to have been described before. In the living parasites examined with the dark-ground illumination he has been able to make out thread-like appendages attached either to the end of the flagellum, or to the posterior end of the body, or both. These appendages are not always present. He was never able to observe them attached laterally to the trypanosomes. The threads are very delicate. They are of constant thickness throughout and are only slightly refractile. The point where the flagellum and the thread-like appendage meet is clearly visible owing to the difference in thickness and the difference in refractility. The threads vary in length, but as the rule the anterior ones are longer than the posterior, and may be as much as twice the length of the body. Occasionally the author observed double or divided threads. The posterior threads as a rule remain extended, but the anterior ones move with the flagellum. In some cases the end of the thread remains attached to the cover-glass and thus fixes the trypanosome to a certain extent.

Apparently the threads possess no power of motion of their own. They are very easily destroyed. In a dying trypanosome they disappear in a few minutes. They first become pale, then granular, and, finally, disappear as if dissolved.

These structures cannot be confounded with those described by WASIBLEWSKI and SENN, or WENDELSTADT and FELLNER, because the bodies seen by those authors occurred at the posterior end of the body only and were observed in stained preparations. Nor can they be confused with the free flagella described by v. PROWAZEK because they are not connected with the blepharoplast.

The author, while unable to form any opinion as to the significance of these appendages, believes that they are composed of the protoplasmic substance.

- (143) NATTAN-LARRIER (L.). **Non-Transmission des Trypanosomiasis de la Mère au Foetus.** [The Non-Transmission of Trypanosomiasis from the Mother to the Foetus.]—*Bull. Soc. Path. Exot.* 1912. Vol. 5. No. 7. pp. 550-556.

Difficulties were encountered in the experiments made in connection with this question owing to the frequency with which abortion followed the inoculation of the females.

Two series of experiments were carried out simultaneously as follows:—

- (1) Experiments to test whether trypanosomes are able to pass the placenta and gain access to the blood of the foetus.
- (2) Experiments to ascertain whether the serum of the foetus acquires any special characters resulting from the infection of the mother.

(1) Twelve experiments in all were made in this connection, eight with guinea-pigs, two with rats, and two with mice.

The following was the technique employed in every case.

If parturition occurred spontaneously the young were immediately killed and their blood used for the inoculation of mice. In the majority of cases, however, the animals were killed before parturition occurred, the abdomen opened, and the vagina and fallopian tubes ligatured. The surface of the uterus was then extensively cauterised, an opening being made with a needle heated to redness. The amniotic fluid was withdrawn with a pipette, the opening in the uterus enlarged, the foetuses withdrawn, and the umbilical cords ligatured and cut.

The foetuses were washed in sublimate and salt solution, the blood being withdrawn from the heart with a pipette. In every case about a dozen drops of amniotic fluid and a similar quantity of blood were used for the inoculation of a number of mice. Finally both the blood and the amniotic fluid were examined microscopically.

Proceeding in this way sixteen mice were inoculated, but the result in every case was negative. The trypanosomes used were: *T. evansi*, *T. brucei*, *T. congolense*, *T. soudanese*, and *T. gambiense*.

The author thinks that before expressing an opinion as to whether this holds true for all species of trypanosomes experiments should be made with *T. equiperdum* since this organism is able to pass through intact mucous membranes. He also suggests that *Schizotrypanum cruzi* might also be capable of causing hereditary lesions.

2. Experiments to ascertain whether the serum of the progeny of infected females acquires any special characters.

In this connection two points have been investigated, viz., whether the serum possesses any protective power, and whether it is capable of causing agglutination.

Five experiments were made to test the serum of the young animals as to the presence of protective substances, the results being controlled by parallel tests with the serum of the mother. In one case both sera exercised a complete protective effect, in two cases the two sera produced a delay in the development

of infection to almost the same degree, and in the other two cases there was no effect whatever. The activity of the amniotic fluid was always found to be inferior to that of the serum.

The author has completed a single experiment only regarding the agglutinating power of the sera, and the result shows that the two sera produce agglutination to practically the same extent.

(144) WOLBACH (S. B.) & BINGER (C. A. L.). **A Contribution to the Parasitology of Trypanosomiasis.**—*Jl. of Medical Research*. 1912. Sept. Vol. 27. No. 1. pp. 83-107. With 6 plates.

The purpose of the work upon which the paper was based was to study the distribution of the trypanosomes in the tissues, and to ascertain the factors concerned in the production of the lesions of trypanosomiasis. This was stimulated by the acquisition of a reliable method of staining trypanosomes in the tissues. The method is as follows:—

1. The pieces of tissue for examination must be taken immediately after the death of the infected animal. They should be cut into slices not more than two millimetres thick and fixed for at least forty-eight hours in a mixture consisting of two parts of saturated solution of corrosive sublimate in distilled water and one part of absolute alcohol. The pieces may remain in this mixture for weeks without harm.

2. The pieces are dehydrated in alcohol, clarified in cedar oil, and embedded in paraffin. The sections should not be more than four microns in thickness, and it is better to have them half that thickness as the stain has poor penetrating power.

3. The paraffin is removed with xylol, followed by graded alcohols and water.

4. Treat with the following solution of iodine for fifteen to twenty minutes: Saturated solution of iodine in ninety-five per cent. alcohol three or four cubic centimetres; seventy per cent. alcohol, one hundred cubic centimetres.

5. Treat with eighty per cent. alcohol until the yellow colour is removed. Wash with distilled water and place in half per cent. aqueous solution of hyposulphite of sodium for ten minutes. Wash in running water for five minutes and rinse in distilled water.

6. Stain in freshly prepared Giemsa solution (60 to 80 drops to 100 c.c. of water) for four to twelve hours. The stain should be replaced twice by fresh mixtures during the first hour. The water used for diluting the stain must be absolutely free from acids.

7. Transfer the sections for differentiation through two changes of colophonium in acetone (15 per cent. solution). Fifteen to twenty seconds generally suffice.

8. Pass the sections rapidly through acetone-xylol (70 to 30), xylol, cedar oil, and mount in cedar oil.

According to the authors this method gives the same effect as that obtained in film preparations.

Two strains of trypanosomes were used during the investigation; a strain of *T. brucei*, originally brought from Uganda, and a strain of *T. gambiense* from the French Congo. Specimens for examination were obtained from four white rats, six guinea-pigs, and three monkeys infected with the *T. gambiense*, and from four white rats and three guinea-pigs infected with the *T. brucei*.

The authors state that they have not been able to make any substantial contribution to the pathology of trypanosomiasis, but they note the development of skin lesions associated with giant cell formation in the guinea-pigs infected with *T. gambiense*.

The following conclusions are drawn.

"In trypanosomiasis the trypanosomes do not remain confined to the blood vessels or the lymphatics. They invade the connective tissue structures of all the organs, the reticular tissue of the lymph nodes and spleen, and the substance of the brain.

"The lesions of trypanosomiasis are due to the presence of the trypanosomes in the tissues.

"The most common form of trypanosome in tissue, and we believe the one most active in the production of lesions, is the flagellate form."

SPIROCHAETOSIS.

- (145) DEUTZ. Ueber Versuche zur Uebertragung von Hühnerspirochaeten auf Mause. [The Transmission of the Spirochaete of the Fowl to the Mouse.]-*Hyg. Rundschau*. 1912. Aug. 15. Vol. 22. No. 16. pp. 1017-1019.

The experiments of LEVADITI and LANGE have shown that the spirochaete of the fowl produces in rabbits inoculated with it a very benign disease only. The author therefore investigated the effect of inoculating mice with this organism. The mice used were inoculated either intravenously into the veins of the tail or intraperitoneally. The results varied in different cases. Whereas in some of the mice spirochaetes could be found in the blood 72 hours after inoculation, in others none could be found only 24 and 48 hours after. As the parasites disappeared from the circulation certain abnormalities of the blood made their appearance. There appeared to be an increase in the number of the blood corpuscles and blood platelets. The leucocytes appeared to be larger, and vacuole formation and amoeboid movements were observed to be far more prominent than normal. Appearances suggested that a process of phagocytosis was going on, and observations made at intervals of 10 to 15 minutes showed that such was the case.

Attempts were made to infect mice from each other by intravenous inoculation, but in no case was this successful for more than two passages. Using mice from 10 to 20 days old, three passages were obtained when the inoculations were intraperitoneal. As a general rule spirochaetes disappeared from the blood within 40 hours after intraperitoneal inoculation. Examination of the peritoneal fluid showed that within the course of the first hour after intraperitoneal inoculation a proportion of the organisms were ingested with phagocytes. Mice that have been inoculated with blood containing the organism fail to react to a second inoculation. If a recovered mouse be inoculated intraperitoneally, and the peritoneal fluid be examined immediately after the inoculation, typical agglomeration of the spirochaetes can be observed, the agglomeration being promptly followed by phagocytosis. The admixture of serum from a recovered mouse, or from one that has been repeatedly inoculated, with fresh actively motile spirochaetes causes an almost immediate cessation of movement. This is not observed if normal mouse serum be used. If the serum be taken from a mouse immediately spirochaetes have disappeared from its blood this phenomenon is not observed, the serum behaving exactly like normal serum.

Spirochaetes that have been passed through mice appear to have lost some of their virulence as indicated by the fact that when such spirochaetes are used to inoculate fowls the period elapsing between inoculation and death is about three times that observed in ordinary cases—namely six days as compared with forty-eight hours.

- (146) KOLLE (W.), ROTHERMUNDT (M.), & PESCHIÉ (S.). **Untersuchungen über die Wirkung von Quecksilberpräparaten auf Spirochätenkrankheiten.** [Investigations into the Actions of Mercury Compounds in Spirochaete Diseases.]—*Deut. Med. Wochenschr.* 1912. Aug. 22. Vol. 38. No. 34. pp. 1582-1585.

The authors' object has been to investigate the therapeutic and toxic effects of various compounds of mercury, and thus obtain data for the preparation of fresh compounds of high value for destroying spirochaetes but of low toxic value for the animal host.

A very large number of preparations have been tested. These are given in the form of a table and the following classes of compounds are included. Soluble and insoluble inorganic compounds of mercury; Soluble and insoluble organic compounds, compounds of the aliphatic and aromatic series being included in each group; Compounds of mercury with albumin and its cleavage products, both soluble and insoluble; Colloidal mercurial compounds.

- (147) LEVADITI (C.). **Intervention de l'Organisme dans la Guérison médicamenteuse des Maladies à Spirilles.** [The Part played by the Body in Recovery from Diseases caused by Spirilla brought about by the Administration of Drugs.]—*Bull. Soc. Path. Exot.* 1912. July. Vol. 5. No. 7. pp. 524-544. With 9 curves.

The author reviews briefly the work already published in connection with this subject and then gives details of a number of investigations upon which he bases the following conclusions:—

The rapidity with which the blood is sterilised varies inversely with the period elapsing between the administration of the drug ("606") and the time at which the crisis of the disease would naturally occur. A relatively rapid recovery may result from the administration of smaller doses of the drug if it be given during the pre-critical period of the disease. This conclusion does not apply, of course, to diseases which do not terminate in a crisis.

The author has found that in the case of rats infected with nagana, in which there is no crisis, the late administration of "606" does not produce more rapid recovery, but that if recovery takes place it is actually slower than when the drug is given in the early stages of the infection.

Two hypotheses may be given to explain the advantage of administering the drug during the pre-critical stage of the disease.

In the first place it may be supposed that, as the disease progresses and the crisis approaches, the vitality of the spirilla becomes

modified in such a way as to render them more sensitive to the microbicidal action of "606." It is known that the organisms present in the blood shortly before the crisis have a greater tendency to agglutinate, and survive *in vitro* for a shorter time than organisms taken in the early stages of infection. In this case the more rapid disappearance of the organisms from the blood would be due to a change in the spirilla and not to any special assistance on the part of the animal. The author does not think that this view can be accepted for the following reasons. If there is an actual modification in the vitality of the parasites one ought to be able to inject a fresh animal with spirilla obtained during the pre-critical period, subject the animal to treatment immediately after, and observe the prompt disappearance of the organism. The author's experiments have shown that this as a matter of fact does not happen, but that a change of animal host is associated with the immediate disappearance of the favourable influence of the pre-critical period.

The second hypothesis, which appears to the author to be the more probable, is that if the rat treated during the pre-critical period recover rapidly it is because the animal has acquired during this period the power of destroying the spirilla more easily. In administering the drug at this opportune moment the crisis is forced on and the host employs its ordinary defensive powers for the destruction of the parasites. It has been shown that in the treatment of spirillosis in the rat there is phagocytosis of the parasites, but also that the same phagocytosis is observed during the crisis which terminates experimental recurrent fever.

It will thus be observed that there is a particular time during the course of the disease at which it is most beneficial to administer the drug.

(148) NOGUCHI (H.). *The Pure Cultivation of Spirochaeta duttoni, Spirochaeta kochi, Spirochaeta obermeiri, and Spirochaeta novyi.*—*Jl. Experimental Med.* 1912. Aug. 1. Vol. 16 No. 2. pp. 199-210. With 2 plates.

The medium employed by the author for the successful cultivation of these blood parasites is prepared as follows. Pieces of fresh sterile tissue, usually rabbit kidney, are placed in sterile test-tubes. A few drops of citrated blood from the heart of infected rats or mice are added, and then about fifteen cubic centimetres of sterile ascitic or hydrocele fluid are poured into the tubes. To some of the tubes a small quantity of sterile paraffin oil was added. The tubes were then incubated at 37° C. It is very important that the ascitic fluid should contain no bile, and should be capable of forming a loose fibrin in the tubes. Fluids which have been heated to 60° C. for half an hour are unsuitable, as also are fluids that have been passed through a Berkefeld filter. The addition of broth or sugar diminishes the value of the medium.

The maximum growth is reached in about a week. The organisms do not multiply in an atmosphere of hydrogen or *in vacuo*, and no growth is obtained at room temperature.

Subcultures should be made by inoculating fresh tubes with a half to one cubic centimetre of culture from the fourth day up to the day of maximum growth. The organisms can be kept for a number of generations operating in this way, and the pathogenicity of the spirochaetes is not lost, although there appears to be a tendency to attenuation after a large number of subcultures have been made.

Growth is slower in the tubes containing paraffin oil.

- (149) NAKANO (H.). Ueber die **Reinzüchtung der *Spirochaeta pallida***. [The Pure Cultivation of *Spirochaeta pallida*.]—*Deut. Med. Wochenschr.* 1912. July 11. Vol. 38. No. 28. pp. 1333-1335.

The author has been successful in obtaining cultures of the organism in impure culture in a number of different media, and it was with the object of purifying such contaminated cultures that the technique described in this paper was devised.

A sterile glass cylinder measuring about 7·5 by 2·5 centimetres is partly filled with sterile horse serum. A bacterial filter is then inserted into the cylinder. Both the cylinder and the filter are closed with rubber stoppers. The apparatus so fitted up is placed on four consecutive days for four hours in a water bath at 58°. On the fourth day it is placed in a water bath at 65° for 30 minutes, the contents being gelatinised in this way. The apparatus is tested by incubating it at 37°. The contaminated culture is introduced into the filter and the apparatus again placed in the incubator at the body temperature.

The author has been able to obtain pure cultures of a number of strains of the spirochaete in this way, including primary cultures. In some cases the colonies have appeared outside the filter on the third day. Immediately colonies are observed subcultures must be made from them because, although the spirochaetes pass through the filter more rapidly than bacteria, the bacteria do eventually escape. A few details are given regarding the appearance of the colonies, and the structure and characters of the cultivated organisms.

- (150) PROCA (G.), DANILA (P.), & STROE (A.). **Sur l'isolement des Spirochètes**. [The Isolation of Spirochaetes.]—*Compt. Rend. Soc. Biol.* 1912. July 26. Vol. 73. No. 27. pp. 235-236.

The method which the authors have found most successful is that of SOWADE slightly modified. Having obtained a mixed culture containing a large number of spirochaetes, subcultures are made in a number of tubes containing pyrogallol serum coagulated in the vertical position at 80° C., the medium being stabbed in the upper third only. The tubes are thus left for 10 to 20 days at 37° C. In the interval the serum beneath the zone of liquifaction is examined to see if it contains motile spirochaetes. If such be found the liquified serum is pipetted off and the tube is broken,

several fresh inoculations being made with particles of serum containing the spirochaetes in pure culture.

In this manner it is comparatively easy to obtain cultures which are pure in the lower half of the serum. Sometimes, however, symbiosis of spirochaetes is encountered. Sub-cultures with the pure spirochaete in pyrogallol serum, in simple serum, or in serum diluted with a solution of gentian always remain sterile. The spirochaetes become immobile after the first 24 hours.

Spirochaetes isolated in the lower layers of pyrogallol serum multiply abundantly when in making sub-cultures the typhoid bacillus is added. The bacillus mesentericus can also be used provided the medium employed be gentian serum. If the bacilli are added after 24 hours, by which time the spirochaetes have lost their motility, no effect is obtained. The simultaneous inoculation enables one to obtain abundant growth in every tube. After ten days spirochaetes are to be found throughout the serum but no actual colonies are formed.

Liquid media such as serum or serum broth are unsuitable. However, abundant cultures can be obtained in liquid serum if ordinary agar in the proportion of one in seven or eight of serum be added, the melted agar being well mixed with the serum at the time of inoculation. The cultures are odourless. The spirochaetes used by the authors were obtained from syphilitic lesions of the vulva.

- (151) SCHERECHEWSKY (J.). *Reinzüchtung der Syphilisspirochaeten.* [The Pure Cultivation of the Spirochaete of Syphilis.]—*Deut. Med. Wochenschr.* 1912. July 11. Vol. 38. No. 28. pp. 1335-1336.

Tubes of sterile horse serum are plugged and capped and then placed in a water bath at 57° C. The temperature of the bath is the gradually raised until it reaches 70° C. at which temperature the serum becomes gelatinised. Immediately this condition is reached the tubes are placed in an incubator at body temperature to determine their sterility. Papules are washed with boracic acid solution and then cut out with curved scissors, care being taken to cut sufficiently deeply. The pieces of tissue are placed in the culture tubes and gently forced down into the depth of the medium, which returns and covers them. The tubes are then capped and placed in an incubator at 37° to 40° C.

The spirochaetes owing to their motility penetrate the medium, while contaminating organisms grow around and above the piece of tissue. On the fifth or sixth day a canal is made through the medium with a sterile capillary tube provided with a rubber teat. This canal reaches down to the piece of tissue, and into it about 3 cc. of 70 per cent. alcohol are ejected. The alcohol is allowed to remain for ten minutes and is then replaced by sterile distilled water, which in its turn is replaced by sterile paraffin oil. The tubes are again capped and incubated for a further five days. A scratch is made on the tube below the level of the now sterilised contamination and the tube broken off. In the medium contained in this part of the tube the spirochaetes are found in pure culture.

LEISHMANIASIS.

- (152) JEMMA (R.). Sulla Leishmaniosi del Cane nei Dintorni di Palermo. [Leishmaniasis of the Dog in the Neighbourhood of Palermo.]—*Pathologica*. 1912. Aug. 1. Vol. 4. No. 90. pp. 466-467.

Reference is first made to a previous publication by the author in which he stated that he had examined 227 dogs in Palermo for Leishmania with negative results. In the present investigation the parasite was found in two dogs. In the first instance the infected dog was found in a house near to one in which there was a child infected with the disease. The parents of the diseased child had had their own dog killed on account of its very poor condition, and it is probable that it also was infected. The diseased dog was examined, preparations being made from the liver and the bone marrow. In both of these parasites were found in large numbers.

In the second instance the owners of the dog could not be traced. Microscopic examination of the liver and bone marrow showed numerous parasites.

Seven dogs in all were examined in the neighbourhood in which there are numerous cases of human leishmaniasis, but in two only was the parasite found.

BASILE was associated with the author in the investigations.

- (153) MARSHALL (W. E.). Further Experimental Investigation into Sudan Kala-Azar.—*Jl. R. Army Med. Corps*. 1912. Sept. Vol. 19. No. 3. pp. 276-280. With 1 plate.

Experimental Kala Azar in the Dog.—Four dogs have been infected, and it has been shewn that the dog can be infected experimentally from the monkey, from another infected dog, and from human cases. Young dogs appear to be more susceptible, and in them the disease runs an acute course. In two experiments attempts were made to transmit the disease from dog to dog by means of the dog flea, but without success. Ticks also failed to transmit the disease in one experiment.

Experiments to determine by what means the disease may be conveyed from monkey to monkey failed, save in one case in which the result was doubtful. In this instance, if there was actual infection, the louse, the flea, and the mosquito may have played some part in the transmission. The author has encountered one monkey that appeared to have a natural immunity.

- (154) NICOLLE (C.) & CONOR (M.). Quelques Expériences Prati-
quées avec le Virus de la Leishmaniose Naturelle du Chien.
Reproduction de la Maladie chez le Singe. [Experiments with
the Virus of Canine Leishmaniosis. Reproduction of the
Disease in the Monkey.]—*Bull. Soc. Path. Exot.* 1912.
June. Vol. 5. No. 6. pp. 351-355.

The virus with which the experiments were made was obtained from YAKIMOFF, it having been found in a dog early in 1911. The lesions presented by the dog were the following: Emaciation,

purulent discharge from the eyes, loss of hair on some parts of the skin, enlargement of the lymphatic glands, thickening of the pericardium, congestion of the bone-marrow. Parasites were found in large numbers in smears from the bone marrow, few in the spleen, which was not enlarged, and none in smears from the liver, kidneys, lungs, or lymphatic glands.

Cultures were obtained, but they were contaminated, and subcultivation failed.

Two fresh dogs were inoculated successfully from the first animal. One of these died accidentally, but was found to be severely infected. No fresh dogs were inoculated from this animal.

The second dog was killed in December, 1911. The animal was fat and showed no gross lesions. On microscopic examination the parasite was found to be scantily present in the spleen, liver, and marrow. Cultures were obtained from the spleen on NNN medium, and subcultures were successful.

From this dog three further dogs and two monkeys were inoculated. In two of the dogs, which were old, the inoculation failed.

The third dog was killed about nine weeks after inoculation. There were no gross lesions, but parasites were found to be numerous present in the spleen, and bone-marrow, scanty in the liver and mesenteric glands, and none were found in the lungs, kidneys and blood. An attempt to inoculate another dog from this one failed.

One of the monkeys, an adult, failed to become infected. The second, which was very young, was suddenly seized with illness and showed symptoms of suffocation about six weeks after inoculation. It was killed on the evening of the same day when in a moribund condition.

Nothing was found to account for the suffocation. The heart was enormously dilated, but there were no lesions either of the myocardium or the valves. The spleen was enlarged.

In smears from the spleen parasites were fairly numerous; both intra- and extracellular forms being encountered. There were very few in the marrow and liver, and none were found in other organs.

Attempts to transmit the infection from this dog to another dog and a monkey failed.

The cultures obtained had up to the time of writing been carried on through twelve generations on NNN medium, and the characters presented were exactly those of cultures of the human parasite.

Two intraperitoneal inoculations with large quantities of culture into a monkey failed to set up infection.

(155) YAKIMOFF (W. L.) & KOHL-YAKIMOFF (N.). *L'Infection des Animaux de Laboratoire par Leishmania infantum* Ch. Nicolle. (Deuxième Note Préliminaire.) [The Infection of Laboratory Animals with *Leishmania infantum*.]—*Bull. Soc. Path. Exot.* 1912. June. Vol. 5. No. 6. pp. 355-357.

Infection of dogs by cultures.—The first dog was twice inoculated intravenously with cultures at an interval of three weeks. About four months later bone marrow was obtained by trephining

and *Leishmania* was discovered. A fortnight later the dog was killed and parasites were found to be fairly numerous in smears from the spleen and bone marrow, but none were found in smears from the liver, lungs, and lymphatic glands. A culture was obtained on NNN medium.

A second dog, inoculated intravenously with four tubes of culture a few days after the first, died on the forty-eighth day. Parasites were found to be present in the marrow in small numbers.

Infection of mice and rats with materials from infected organs.—The results of the inoculation of eleven mice and one rat are given. From these it is seen that in every mouse save one parasites were found in smear preparations from various organs. In one mouse and the rat microscopical examination was negative, but in both instances cultures were obtained from fragments of the spleen.

(156) WENYON (C. M.). *Some Recent Advances in our Knowledge of Leishmaniasis.*—*Jl. London School of Trop. Med.* 1912. March. Vol. 1. Part 2. pp. 93-98.

Dogs have been found liable to Kala Azar in practically all the great centres of the infantile form of the disease—Tunis, Algeria, Lisbon, Malta, Sicily, Rome, and Greece. It has been suggested by NICOLLE that the dog, in which the disease is mild and chronic, may act as a reservoir for the virus, which is transmitted to man by some biting arthropod. BASILE claims to have proved that the transmission is effected by the dog flea (*Ctenocephalus canis*) and the domestic flea (*Pulex irritans*). His experiments were carried out on the following lines. First, healthy dogs introduced into houses where the disease existed became infected: fleas from infected houses in Sicily were taken to Rome and placed upon healthy, isolated dogs, which contracted the disease: dog fleas fed upon the spleen-juice of an infected dog were found to become infected with the cultural form of the organism. The gut-contents of fleas are infective to healthy dogs. GABBI failed to confirm these results and is inclined to incriminate the mosquito on the grounds of positive results obtained by FRANCHINI. MARSHALL, in the Sudan, has observed infection of a healthy monkey living in a cage with diseased ones. The organism is inoculable to the dog and monkey, and successful inoculations have been made into guinea-pigs, rats and rabbits. In the monkey the disease resembles that seen in children. A few experiments have been made in India to inoculate dogs but without success.

A certain amount of evidence has been adduced by NICOLLE and MANCEAUX that dogs that have recovered are immune.

Cultures of the parasite of Oriental sore, first obtained by NICOLLE on blood-agar, retain their virulence. Most observers are agreed that the house-fly may play some part in the transmission of the disease, and ROW has found that the parasite may remain infective for monkeys for three hours after having been taken up by the fly. Wenyon has found that bed-bugs and

Stegomyia fasciata would feed on sores and take up parasites, and that in their guts the parasites develop into flagellate forms as they do in culture.

DONOVAN examined more than a thousand pariah dogs in India but failed to find infection, but possibly such dogs are immune.

RABIES.

- (157) BOUFFARD (G.). Sur l'Existence de la Rage Canine dans le Haut-Sénégal et le Niger. [Canine Rabies in Senegal and the Niger.]-*Ann. Inst. Pasteur.* 1912. Sept. 25. Vol. 26. No. 9. pp. 727-731.

While there is no record of the occurrence of rabies in the human subject in the French colonies in West Africa, there would appear to be a disease of the dog the symptoms of which are strongly suggestive of rabies. It also appears to be certain that the bite of such dogs is fatal to other dogs, but without effect upon human beings.

The first animal suffering from this disease came into the author's possession in 1906. Unfortunately it escaped before any examination could be made, but not before it had bitten two other dogs belonging to the medical establishment.

One of these dogs sustained only a small bite on the paw which healed promptly, the dog remaining healthy for a year. The other dog was badly torn above the left eye. The wound was dressed and within a week healing was apparently complete, but on the 11th day the animal began to show peculiar symptoms. There was loss of appetite and an absence of desire to move, and throughout the following day the animal remained in a corner. About 6 o'clock in the evening it got up and began to rush about its enclosure in a state of great excitement. The author kept the animal under observation for an hour and noticed that from time to time it appeared to reel and to be uncertain in the movements of its hind legs. The next morning it was found dead in its cage.

Nothing interesting was found at the post-mortem save that the stomach was empty.

A fragment of the medulla was used for the inoculation of a rabbit, but the rabbit died on the fifth day from some accidental cause and its medulla was found to be non-infective for other rabbits. The strain was thus lost.

The following year while investigating sleeping sickness on the Bani River the author encountered a chief who informed him that there was a very fatal disease amongst dogs in his district. The affected dogs appeared to be mad, their bite was fatal to other dogs, but did not appear to cause any harm to human beings. This chief had no knowledge of such a disease occurring in man. Unfortunately the author was not in a position to inoculate animals and get the strain.

In August, 1909, the author came into possession of a dog at Kouloubah showing all the symptoms of dumb rabies. The animal was killed at once and its medulla used for the sub-dural inoculation of a rabbit.

For fifteen days this rabbit appeared to be in perfect health but on the sixteenth day it was found lying in its cage and when made to move it was observed that there was evidence of paralysis of the hind-quarters. The next day the paralysis was complete and the animal lay on its side showing marked dyspnoea. Death took place the same evening.

The medulla of this rabbit was used for the inoculation of two fresh rabbits.

During the next eight months thirteen passages were made, the period of incubation of the disease averaging about fifteen to twenty days. In two cases the period was twenty-five and in one case thirty-eight days.

The symptoms in every case were those already mentioned and in three cases there were additional symptoms of a furious type which made their appearance from twelve to twenty-four hours before death.

At each passage two or three animals were used and it was observed that about one animal in every six appeared to be refractory.

The author's successor continued the inoculations, but used one rabbit only at each passage with the result that the strain died out at the twenty-first passage, this animal remaining healthy.

This rabbit was used five months after the inoculation for a vaccine lymph control and two days later became paralysed and died shortly after.

Three further passages were made, the period of incubation being ten days in each of the first two passages but the third animal inoculated proved refractory and the strain was again lost.

(158) HARRIS (D. L.). *Recherches sur les Propriétés du Virus Rabique conservé à l'Etat Sec.* [The Properties of Rabic Virus preserved in the Dry Condition.]—*Ann. Inst. Pasteur*. 1912. Sept. 25. Vol. 26. No. 9. pp. 732-735.

The author's object was to devise some method of preserving the rabic virus which would obviate the difficulties attaching to the method devised by PASTEUR, in order that a quantity of material might be kept at hand in places where vaccination is required only occasionally.

It has been shown by a number of authors that the virulence of the infective material is retained if a thin layer be made and then dried rapidly by means of sulphuric acid in vacuo. The amount of material that can be preserved in this way is too small to be of any value for vaccination purposes.

In conjunction with SHACKELL the author has described a method of preserving entire brains and cords in which the materials are frozen before being dried. Briefly the plan is as follows:—

The brain is first frozen with a mixture of ice and salt, and then without allowing it to thaw it is dried in vacuo with sulphuric acid.

Numerous experiments showed that about 1 per cent of the original virulence was retained in this way.

The authors believe that the desiccation of the cords according to PASTEUR's procedure causes a progressive concentration of salts and other substances normally contained in the cord and that the destruction of the virus is proportional to this concentration.

During his experiments HARRIS has found that the degree of virulence retained in a cord preserved in this way depends upon the rapidity and completeness with which the material is frozen, and that it is possible to preserve from 30 per cent to 50 per cent. of the virulence. The method is as follows.

The brain or spinal cord is ground up in a porcelain mortar and water is added drop by drop until a thick homogenous paste is obtained. A little carbonic acid snow is then added slowly and with constant stirring in order to avoid freezing the material into a solid mass. When freezing is complete the material is as fragile as glass and can be easily pulverised. It is necessary to add a little of the snow from time to time to prevent thawing.

The mixture is then immediately transferred to a cold vessel and placed in the bottom of an exsiccator which has been previously embedded to half its depth in a mixture of ice and salt at a temperature of -18°C . There is placed in the upper part of the exsiccator a vessel containing sulphuric acid in such a manner that there is free circulation of air between the vessel containing the cord and the exsiccator. Solidification occurs if the acid be placed too near the mixture. There should be a vacuum of 2 mm. of mercury, and the exsiccator should be gently agitated from time to time in order to mix thoroughly the absorbed water with the acid.

A brain can be desiccated in this manner in from thirty-six to forty-eight hours, but towards the end of the operation the temperature must not pass -10°C .

The dried product forms a very light hygroscopic powder which absorbs water from the air up to 3 per cent and then becomes soft, and within a few hours loses all its virulence. To protect the material from moisture it is sealed up in tubes.

It has been shown in numerous experiments that 0.00002 g. of cord dried in this way produces rabies in rabbits on the 6th day. Half this quantity kept at $8-10^{\circ}\text{C}$. for a month and protected from light is also capable of setting up the disease. After two months the virulence is diminished by half.

Phosphoric acid is less satisfactory than sulphuric acid for the desiccation.

Rabbits and dogs can be rapidly immunised with this material by HÖGGES' method, and the author has immunised dogs with cords preserved for three months.

Further experiments are in train to ascertain the effects of certain chemical substances, temperature, light, etc. on the loss of virulence.

The advantages claimed for the method are, that the powder can be easily weighed, its properties can be determined exactly, and that it is relatively permanent.

HELMINTHS.

(159) CONNAL (A.). Some Nematode Worms from Lagos.—*Jl. London School Trop. Med.* 1912. July. Vol. 1. Part 3. pp. 229-237.

The six species of worms described were found in a collection made from various small wild mammals, birds, and reptiles, by BEALE-BROWN in Lagos.

Strongyluris streptoesophageus n. sp. Found in large numbers in the stomach and intestine of the common West African lizard, *Agama colonorum*.

Male. Length 8.6 mm. Breadth 0.44 mm. Body tapers gradually to the head end. Tail perfectly straight. Cuticle smooth. Mouth provided with three simple lips. The oesophagus shows a sharp kink near the anterior end, and behind this the tubes show three bends. There is a well-marked bulb. There are slender subcuticular papillae along the whole length of the body. The anus is quite close to the tail. The spicules are long, equal in length, and tapering. There is a sucker ventrally placed 0.76 mm. from the tail. There are three pairs of pre-anal and six pairs of post-anal papillae.

Female. Length 9.1 mm. Greatest breadth 0.5 mm. Vulva 3.33 mm. from the tip of the tail. Oviparous, the ova measuring 0.06 by 0.03 mm. The sucking disc is absent. Anus 0.2 mm. from the tip of the tail. The species appears to be closely allied to the *Strongyluris brevicaudata*.

Filaria yaba. n. sp. Found in large numbers in the thoracic cavity of *Centropus senegalensis*.

Male. Length 19 mm. Greatest breadth 0.6 mm. Extremities taper somewhat abruptly. Tail bluntly pointed and coiled at least 1½ times. Skin smooth. Mouth simple with a minute papilla on the outer aspect of each of the three lips. Oesophagus somewhat bottle-shaped. Anus about a tenth of a millimetre from the tail. Spicules short and boomerang-like in shape. One pair of pre-anal and four pairs of post-anal papillae, all small.

Female. Length 23-33 mm. Greatest breadth 0.8 mm. Both ends bluntly pointed. Vulva about 1 mm. from the head. Ovoviparous. Embryos much coiled. Anus nearly terminal.

Triplotriaena falconis, n. sp. Found in large numbers in the thoracic cavity of a hawk (sp.?).

Male. 30.1 mm. Greatest 0.46 mm. Tapers at both ends. Cuticle finely striated. Mouth has three simple lips devoid of papillae. Oesophagus slender. Spicules of unequal length. Larger one straight, blunt pointed, and tapering. Shorter one corkscrew-like with four distinct twists, more pointed than the long one and slightly narrower. No anal papillae. Anus nearly terminal.

Female. Length 55 mm. Greatest breadth 0.6 mm. Vulva very close to the anterior extremity. Ovoviviparous. Anus nearly terminal.

Ascaris rosarius n. sp. Found in the stomach of a small grey heron.

Male. Length 26 mm. Greatest breadth 0.6 mm. Anterior end of the worm cylindrical and narrow for a distance of 0.7 mm.

then broadens abruptly. Posteriorly the body tapers rapidly to a sharp point. Cuticle strongly striated, and numerous regularly arranged refractile dots give a beaded appearance to it. Tail coiled ventrally. Mouth provided with six fleshy lips, three large and three small placed alternately. Distinct collar of cuticle around the base of the lips. Oesophagus gradually widens, and there is a blind prolongation of it about 0.5 mm. in length running alongside the intestine. There is a blind prolongation of the intestine forwards alongside the oesophagus. Anus 0.22 mm. from the tip of the tail. Three pairs of post-anal papillae. Spicules long, equal in size and shape, and have a flanged appearance. Numerous pre-anal papillae.

Female. Length 30 mm. Greatest breadth 0.7 mm. Vulva 1.5 mm. from head. Oviparous. Shell of the eggs thick.

Spiroptera deflecta. n. sp. Found in the stomach of the palm squirrel.

Male. 19.7 mm. Greatest breadth 0.6 mm. Body tapers to a fairly sharp point anteriorly. The anterior portion bent laterally at a distinct angle. Tail tapers more quickly than the anterior end, and shows a marked curve. Cuticle finely striated. Mouth has six small lobular lips. Oesophagus distinctly bulbous at its junction with the intestine. The opening of the oesophagus into the intestine is furnished with three valvular flaps depending from a collar-like thickening. Anus situated 0.4 mm. from the tail. Spicules short and broad, and unequal in size and shape. The dorsal part and the tip of each is thickened, while the thinned and broadened ventral portion presents a frescoed appearance. There is a curved accessory piece. At the tail end on the ventral aspect there is a well-marked ala nearly 2 mm. in length. Five pairs of pre-anal and three pairs of post-anal papillae.

Female. Length 26 mm. Vulva situated about the middle of the body. Anus 0.3 mm. from tail.

Echinorhynchus centropi, PORTA, 1910. Found in the intestine of *Centropus senegalensis*.

Male. Length 29 mm. Greatest breadth 1.2 mm. Body cylindrical but somewhat swollen towards the anterior part. Tail rounded. Cuticle smooth. Rostrum nearly globular. Hooks arranged alternately nine and ten in a row longitudinally, and twenty-six in a transverse row.

The five posterior hooks in a row of nine are smaller than the anterior four. The cirrus-pouch measures 3.6 mm. in length.

Female. Length 48 mm. Greatest breadth 1.5 mm. General characters like those of the male, but the posterior end of the body has a finger-like prolongation 0.34 mm. in length. Oviparous. Eggs thick-shelled and rather elongated.

- (160) INNES (J. A.). *Gastrothylax bubalis*, n.sp. With a Few Notes on the Genus *Gastrothylax* (Poirier).—*Parasitology*. Sept. 1912. Vol. 5. No. 3. pp. 217-226. With 8 text-figures.

The parasite was found by BROWN, British Central Africa, in the stomach of a Rhodesian hartebeest, and about one hundred and twenty specimens were secured.

The parasites were examined in serial sections, and it was found that sections, 10 microns thick, could be obtained if the process of embedding in paraffin were carried out rapidly. If the process occupied more than two hours the specimens were too brittle to cut well. The best results were obtained by staining the parasites in bulk with MAYER's paracarmine for three days, and then differentiating in several changes of 70 per cent. alcohol to which a little 1 per cent. solution of ammonium chloride had been added. Imperfect fixation made it impossible to study the finer histological details. In tracing the ducts rough reconstructions were made.

The generic characteristic of the genus is the large atrium opening anteriorly by the atrial pore.

External anatomy. The parasite is conical in shape with a blunt anterior end. It measures from eight to ten millimetres in length and from three to four in diameter at its thickest (posterior) part. The parasite may be either expanded or contracted, and in the latter case the surface is covered with wrinkles. The posterior sucker in the expanded specimens occupies practically the whole of the posterior part of the parasite, but in contracted forms its diameter is about one-third of that and it is retracted somewhat. The mouth opens anteriorly exactly at the apex of the cone and is surrounded by a mass of muscular tissue. Oral papillae are present.

The transverse opening of the genital atrium opens a little behind the mouth on the mid-ventral line, and it is lined with papillae.

The ventral side of the parasite is slightly concave or straight, the dorsal side being convex.

Internal anatomy.—The alimentary canal consists of a terminal mouth, a muscular pharynx, a short oesophagus which terminates in two blind pouches passing down on either side of the atrium, dorsal to the testes. The gut branches show varied convolutions. The atrium is large and expands posteriorly where it terminates just above the anterior boundary of the testes. It is triangular in cross-section, the apex of the triangle being ventral. The physiology of the organ is not known but it is possibly used as a receptacle for ova. It opens to the exterior on the ventral surface just behind the mouth, and in this region it also receives the genital opening. The reproductive system is almost entirely included between the base of the atrium and the posterior sucker.

The male system consists of a pair of large round testes, which do not extend close to the body-wall of the parasite, nor into the atrial cavity, two vasa efferentia, and a single long vas deferens opening along with the vagina at the genital aperture. The vas deferens and the vagina run dorsal to the atrium.

The female system consists of ovary, shell gland, oviduct, uterus and vagina. The shell gland and the ovary are very close together, and the duct from the shell gland almost immediately joins that of the ovary. These organs are situated between the testes. The ova measure about one hundred and ten by sixty microns.

The excretory vesicle which is very irregular in outline lies between the shell gland and the posterior sucker, its canal is short and does not unite with Laurer's canal.

The muscular tissue of the parasite appears to be concentrated about the suckorial regions.

The parasite is believed to be non-pathogenic.

Notes on the genus Gastrothylax.

An outline of the historical details connected with the genus is given, and also a description of the general features characterising the genus. Finally the distinguishing characters of the ten species comprising the genus are tabulated.

(161) JOWETT (W.). **Nodular Intestinal Disease of Cattle.**—*Journ. Comp. Path. & Ther.* 1912. March. Vol. 25. No. 1. pp. 15-22. With 5 figures.

The investigations are based upon the examination of material that the author has obtained from cattle during post-mortem examinations. He has frequently encountered the disease in cattle in South Africa. The disease is characterised by the formation of nodules in the sub-mucous coat of the intestine, the nodules varying in size from that of a pin's head to that of a pea. The lesions may be scanty or very numerous. It is only rarely that there is any ulceration of the mucous membrane over the nodules, but there may be considerable thickening. The lesions are more frequently encountered in the small than in the large intestine. The colour of the nodules varies with age. The young lesions are as a rule haemorrhagic, the medium-sized are either black or white and black, and the larger or older nodules are either uniformly white or yellowish, and they may have greenish yellow contents. The oldest lesions are for the most part fibrous, and contain caseous or calcareous material.

It is difficult to demonstrate the parasite which is responsible for the condition in any save young lesions. The causal parasite is the embryo of a nematode worm which measures from two to three millimetres in length.

In the nodules the worm is in the embryo stage, but after ecdysis, by which the worm is converted into an immature adult, it escapes into the lumen of the gut. The adult worms measure about two centimetres in length. In the intestine the worms become sexually mature and copulate. The eggs show segmentation while still in the uterus of the female, as is also observed in the closely related parasite of man.

The eggs are passed out and under suitable conditions undergo processes of development leading to the formation of embryos which gain access to a bovine host with either food or water.

It has been shown by MAROTEL that the causal parasite in an oesophagostome, and from the more recent researches of GUILLÉ, MAROTEL, and PANISSET it would appear that the worm responsible for disease in cattle is a distinct species and the name *Oesophagostomum biramosum* has been given to it. The

characters by which this worm is distinguished from the *O. radiatum* are: (1) the possession of a cervical swelling, somewhat hour-glass-shaped but non-vesicular; and (2) two distinct branches to each of the posterior ribs in the caudal bursa of the male, the outer division of each main branch of the dorsal ray being better marked and somewhat longer than is the case in *O. radiatum*.

PLAGUE.

- (162) DUJARDIN-BEAUMETZ (E.) & MOSNY (E.). *Évolution de la Peste chez la Marmotte pendant l'Hibernation*. [Plague in the Marmotte during Hibernation.]-*Compt. Rend. Acad. Sci.* 1912. July 22. Vol. 155. No. 4. pp. 329-332.

It has long been known that in Transbaikal and Mongolia there are persistent plague centres, and that trappers contract the disease from marmottes and tarbagans in these areas. It appears that the marmotte, like the rat and other rodents, acts as a reservoir for the plague virus.

Until comparatively recently it was believed that in countries where the rat is the transmitting agent, the disease persisted in that animal in a chronic form. It is now known, however, that such is not the case, and that for the survival of the virus there must be a sufficient number of rats to assure the continuous transmission of the disease from rat to rat.

In Mongolia where the winter is severe there could be no question of the virus persisting outside the animal body because it is very susceptible to external influences. It therefore had to be supposed that the virus was maintained either in the flea or in the marmotte, since rats are not found in these plague areas.

The experiments of GAUTHIER and RAYBAUD have shown that the bacillus can retain its vitality and virulence in the stomachs of fleas for 45 days in an ice chest, but it was not shown whether such fleas were capable of transmitting the disease by biting after such a period.

It has already been shown that hibernating marmottes resist infection with tuberculosis, and that trypanosomes fail to infect provided a period of five days elapses before hibernation terminates. In some experiments carried out by WURTZ marmottes were inoculated with plague, but owing to the conditions being unfavourable for hibernation the animals awoke and death occurred some days after inoculation.

The authors have made three experiments with marmottes captured during hibernation. The three animals were kept together at a constant temperature of 5-10° C., and were protected from all stimuli likely to wake them.

The first animal was inoculated subcutaneously with virulent plague virus. It was subjected to handling on a number of occasions causing it to wake, but the animal survived till the 61st day having lost about 12 per cent. in weight during the interval.

The second animal was inoculated in a similar manner, but was not handled more than was absolutely necessary. This animal survived 115 days and died after waking at the normal time.

The third animal was kept as a control to show the susceptibility of marmottes to the virus of plague in the waking state. This animal died two and a half days after inoculation.

The possible explanations are that either the virus persists without multiplication, or that multiplication is very much restricted during hibernation, and becomes very rapid immediately the period of hibernation terminates.

(163) MATSUO (K.). Gleichzeitiges plötzliches Auftreten von Pestfällen bei Menschen und Eseln in demselben Gehöft. [The Simultaneous and Sudden Occurrence of Cases of Plague in Man and Donkeys.]—*Centralb. f. Bakt.* 1. Abt., Orig. 1912. Aug. 10. Vol. 65. Nos. 6-7. pp. 417-423. With 1 plate.

The outbreak of pneumonic plague which occurred in Northern Manchuria in October, 1910, and which lasted till April, 1911, was of extraordinary severity and claimed more than 40,000 victims. The author was stationed at Mukden in January, 1911, for duty in connection with the outbreak. Towards the end of February information was received that a donkey had died of plague in Fushun, and shortly after a dog was found to have died from the disease in Changchun.

When the outbreak was beginning to die out the author accidentally encountered a case of the disease in a donkey, and by following up the case discovered a further instance of infection in the donkey in which nearly every man and donkey in a particular building became infected one after the other. The circumstances of the case were as follows:—

A miller in Mukden employed eleven men and kept twelve donkeys. In March one of the men died, plague being particularly prevalent in Mukden at the time. The employer attempted to conceal the death, and placed the body in a loft where it was subsequently found, and plague was proved to have been the cause of death.

Soon after the man's death one of the donkeys became ill, losing its appetite and coughing severely. The animal was sold shortly after and its purchaser could not be traced. After a short interval another man died. Subsequently seven donkeys became infected one from another. Of these three were sold and four died. Two healthy donkeys were sold at the same time as the diseased ones, but none of the animals nor the purchaser could be traced. One of the dead donkeys was removed and buried. Before the other three dead donkeys could be removed the miller and all his employees were placed in the isolation hospital, the donkeys being afterwards buried.

The two remaining animals were isolated and remained healthy. The miller and seven of the nine men died of plague.

The author received instructions to examine the three donkeys buried. The following lesions were found in the first of the three: The brain was putrid but showed congestion, as did also the medulla, there were no adhesions of the pleura, but the lungs were congested, and there was distinct infiltration of the upper border of both lungs. The heart was hypertrophied, liver three times the

normal size, spleen reduced in size with numerous wrinkles, stomach normal, hyperaemia of a portion of the large intestine with petechiae, kidneys normal.

Only the lungs were removed from the other two carcasses, and there was marked infiltration of the borders of the lungs and distinct hepatisation of the right lung.

Fluid was obtained by puncture of the lungs and from the upper air passages and bacilli resembling the plague bacillus were found in addition to numbers of other bacteria—principally streptococci. Smear preparations were made from the various organs and stained by Loeffler's method. Bacilli were found to be numerous present in smears taken from the lungs, scanty in smears from the liver, and absent in smears from the heart-blood, spleen, brain, and kidneys.

Nine mice were inoculated with materials taken from various organs, and practically all died in 40-45 hours, a single one surviving till the 72nd hour. The organisms isolated from the mice agreed exactly in cultural and staining characters with the bacillus of plague.

Owing to the impossibility of getting experimental animals only three experimental inoculations could be made to test the virulence of the bacilli cultivated from the mice. Control inoculations were made with sputum from a case of plague and the results were in absolute agreement, a guinea-pig and a mouse inoculated in each instance with 1/100,000 of a loopful dying in 75-90 hours. Plague bacilli were demonstrated in the blood, liver and spleen of the experimental animals. Two further guinea-pigs inoculated with 1/1,000,000 of a loopful were still healthy some months after.

Immune plague serum (horse) was used to compare the agglutination of the organisms isolated from the donkeys with controls of plague bacilli. Here again the results were in absolute agreement, the serum causing agglutination with both strains of organisms in dilutions up to 100 but not higher.

MISCELLANEOUS.

- (164) LHERITIER (A.), FLEURY (A.), & TRIBOUT (A.). *Moutons Algériens et Bactériologie Charbonneuse*. [Algerian Sheep and the Anthrax Bacillus.]—*Bull. Soc. Path. Exot.* 1912. June. Vol. 5. No. 6. pp. 336-339.

After a brief reference to an outbreak of anthrax among some cattle and to one case in a sheep, the authors give a short review of the experiments carried out by CHAUVVEAU which indicated that Algerian sheep are more or less resistant to the bacillus of anthrax as it occurs in Europe.

Experiments were made with the object of comparing the virulence of the bacillus obtained from an Algerian sheep dead of the disease with that of a strain of the organism obtained from Europe.

In a preliminary experiment it was found that whereas the Algerian strain proved fatal to a guinea-pig in 33 hours, the European strain did not cause death till the 40th hour, the guinea-pigs used being of the same weight, and a similar dose being administered in each case.

In the second experiment Algerian sheep were used. These were divided into two lots of five. One batch was inoculated with the Algerian strain and the other with a French strain. In each series four were inoculated subcutaneously and one intravenously, the doses being strictly comparable.

The whole of the sheep inoculated with the Algerian strain died, while the whole of those done with the French strain recovered.

The sheep which had resisted inoculation with the French virus were inoculated three months later with a large dose of the Algerian strain to ascertain whether they had acquired any immunity. One fresh sheep was inoculated at the same time as a control. The control sheep died, but none of the others showed any symptom save a slight elevation of temperature.

- (165) **LEBOEUF (A.). Existence de *Lepra murium* (*Lèpre des Rats*) en Nouvelle-Calédonie.** [Leprosy of the Rat in New Caledonia.]—*Bull. Soc. Path. Exot.* 1912. July. Vol. 5. No. 7. pp. 463-465.

Having given a brief review of the literature the author states that he has found three infected rats out of ninety-nine examined. He agrees with other investigators that the bacillus found is easily distinguishable from the bacillus of HANSEN. The organism is longer, as a rule stains uniformly, is slightly curved, frequently shows a spherical swelling at one end, and shows no tendency to collect into rounded masses.

The three rats were all adults and appeared to be in perfect health.

In the first acid-fast bacilli were encountered in the axillary and inguinal glands on the left side, and a few in the apex of the left lung.

The second rat had very large axillary and inguinal glands on both sides of the body and bacilli were very numerous present.

The third rat showed fairly numerous bacilli in the left inguinal gland, and they were not scanty in the left axillary gland and in both glands on the right side.

The author points out that in one animal only was there any evidence of invasion of the viscera, and that in the same animal the glands were less seriously affected than in the others.

- (166) **Distribution, Etiologie et Prophylaxie de la Fièvre Ondulante.** [Malta Fever.]—*Bull. Off. Intern. d'Hyg. Pub.* 1912. July. Vol. 4. No. 7. pp. 1180-1211.

This paper gives a brief review of the work done in connection with this disease up to date and deals with the following points: Preliminary considerations, including synonyms; Historical; Distribution; Symptomatology; Prognosis and Evolution; Epidemiology; Etiology and the Biological Characters of the causal organism; and, finally, Prophylaxis. From the veterinary point of view the principal interest centres in the details that are given regarding the extent to which goats are affected in various places,

and the occurrence of infection in animals of other species than the goat. In connection with the occurrence of infection in goats the following table is given:

| Authors. | Place. | Percentage of goats infected. |
|------------------------------|------------|-------------------------------|
| ZAMMIT and HORROCKS | Malta | 50 |
| SERGEANT | Algeria | 3.4 |
| NICOLLE and CONSEIL | Tunis | 30.7 |
| CONOR and HUON | Marseilles | 34.2 |
| AUBERT, CANTALOUBE & THIBAUT | Gard | 29.0 |
| SHAW | Malta | 9.9 |

The goat is far more frequently affected than other species of animals, but epidemics are not all unknown in such animals.

The sheep appears to be next most commonly infected after the goat, and as in the goat abortion is very commonly observed.

The organism has been isolated from the mule, donkey and horse in Algeria. A number of authors have recorded its occurrence in cattle, and a number of birds are susceptible. The susceptibility of the rabbit is not yet fully established.

- (167) BEVAN (Ll. E. W.). **Ephemeral Fever, or Three Days Sickness of Cattle.**—*Veterinary Jl.* 1912. Aug. Vol. 68. No. 446. pp. 458-461.

This paper contains a brief summary of what is known regarding this disease, emphasising some points by reference to particular cases. The disease is remarkable in that outbreaks occur in areas far removed from each other, and between which there has been no interchange of stock. The disease attacks animals of all ages, conditions, and sexes, and animals immune to red-water, anaplasmosis, and Coast fever possess no immunity. The onset of the disease in a herd is sudden, and within a day or two a number of animals may be affected. In Cape Colony the disease has been observed in stable animals. It is known that the virus of the disease is present in the blood of a sick animal, and proof has been furnished that it can be transmitted to other animals by blood inoculation. The period of incubation in such cases is two or three days and the attack is followed by immunity which lasts for about six weeks. The blood of a recovered animal does not transmit the disease. It has been suggested that the disease is insect transmitted.

Typical cases run their course in about three days, but in complicated cases the period of illness may last for weeks. Acute cases have been known to recover within twenty-four hours. The condition must not be confused with "Lamziekte" which is caused by eating *Crotalaria burkeana*.

The stiffness may be observed in any or all the legs, and it may pass rapidly from one limb to another. The muscles of the neck or the back may be involved. Very often ropy saliva hangs from the lips and discharge pours from the nose. The tissues around the eyes may be very swollen. There is marked elevation of temperature, and constipation is frequently observed.

The mortality is low and it is difficult to say what are the lesions proper to the disease.

- (168) SCHELLHASE (W.). Ein Beitrag zur Kenntnis der ansteckenden Lungenbrustfellentzündung der Ziegen in Deutsch-Ostafrika. [Contagious Pleuro-pneumonia of the Goat in German East Africa.]—*Zeitschr. f. Infektionskrankh., Parasit. Krankh., u. Hyg. d. Haust.* 1912. Vol. 12. No. 1. pp. 70-83.

The author, who carried out his investigations in the field without laboratory equipment, arrives at the following conclusions:—

1. The disease is transmissible to healthy goats by intrapulmonary inoculation with lymph obtained from the lung tissue of a diseased animal.

2. The disease is not transmissible by subcutaneous inoculation with the same material.

3. The disease cannot be transmitted to calves by intrapulmonary inoculation.

4. Intrapulmonary inoculation fails to transmit the disease to sheep, but the inoculation causes a localised inflammation of the lung tissue.

5. Subcutaneous inoculation of goats with lung lymph causes a local and a general reaction. The disease cannot be transmitted by intrapulmonary inoculation to sheep so treated.

6. Smears from the lung tissue of natural and experimental cases show large numbers of coccus-like bodies which are easy to stain, but the significance of which could not be determined.

- (169) BRIDÉ (J.), NÈGRE (L.), & TROUETTE (G.). Recherches sur la Lymphangite Épizootique en Algérie. [Epizootic Lymphangitis in Algeria.]—*Ann. Inst. Pasteur.* 1912. Sept. 25. Vol. 26. No. 9. pp. 701-726. With one plate.

The authors deal at some length with the clinical aspects of the disease before passing to the consideration of the pathological anatomy of the condition and the causal organism.

Examination of an excised corded lymphatic vessel shows that in the smallest nodules the contents are of a greyish red colour, whereas in the larger and older lesions they approximate more and more to pus in appearance.

Microscopic examination of one of the smallest nodules shows that the contents appear to consist of a colony of cryptococci, many of which are multiplying; but very few leucocytes are to be seen.

On examining larger lesions it is found that leucocytes have gained access to the mass of parasites and that the number of organisms showing evidence of multiplication is smaller. It is further observed that a number of the organisms have been ingested by phagocytes.

The organism can be stained by GRAM's method, but a large proportion of the elements are decolourised. Better results are to be obtained by CLAUDIUS' method, but both the gentian violet and the picric acid should be allowed to act for an hour and chloroform should be used as the decolourising agent. Good results may also be obtained with the toluidine blue method of DOMINICI and with GIEMSA's solution.

In sections it can be seen that the organisms invade the whole of the lymphatic vessel and a certain number are to be found free among the proliferating connective tissue cells.

The organism is not a parasite of the white corpuscles and the occurrence of elements within leucocytes is simply evidence of phagocytosis.

In the opinion of the authors the view that the cryptococcus is a protozoal parasite is a mistaken one, this view having been based upon the examination of lesions in too advanced a stage and in which phagocytosis was active. They state that when a lesion has become appreciable to the touch in a living animal it is already invaded by leucocytes and phagocytosis has commenced.

The disease must be considered as a local affection which spreads in the centripetal direction along the course of the lymphatics. The organisms rarely or never pass the glands at the entrance to the chest or the abdomen to determine lesions in the internal organs.

A brief review of the opinions expressed by various authors as to the nature of the organism is given. The authors disagree with the view that the double contoured envelope in an artefact, and that lemon-shaped forms are rare. They further disagree with the explanation offered by the partisans of the protozoal theory that budding forms represent two individuals joined together.

In support of their view that the parasite multiplies by a process of budding they state that in the young lesions described numerous parasites are to be found with daughter cells attached to them and showing a gap in the capsule through which the daughter cell is herniated. They further state that parasites may be observed having daughter cells of the second generation also attached to them.

The authors express no opinion as to the nature of the cell contents.

A large number of attempts have been made to cultivate the organism on the most diverse media both of animal and vegetable origin, but without success. In a few cases the authors have thought that there was a larger number of budding forms in certain media than in the seed material, but they have never obtained obvious growths, nor have subcultures ever proved successful.

This failure to cultivate the organism is not evidence of its protozoal nature, for *Leishmania*, to which some authors believe that the organism approximates, can be easily cultivated.

The authors believe that the causal organism is of the nature of yeast and with this idea carried out "deviation of the complement" experiments.

In the first instance the presence of sensitiser was sought in the serum of affected animals using a dilution of cryptococci in salt solution as antigen. The results indicated that the serum of infected animals contains a sensitiser for the cryptococcus.

In the second series of tests a culture of a known blastomycete was used in the place of the dilution of cryptococci as antigen.

The experiments were repeated with several kinds of yeast and with the sera of diseased and normal horses. The results obtained reproduced those obtained in the first series of experiments, positive results being obtained with a rice yeast.

In a third experiment it was found that a bacterium such as the *B. coli* in place of the yeast gave negative results.

A test was then made as to whether the cryptococcus or the yeast was capable of fixing any other sensitiser than that contained in the serum of an animal infected with epizootic lymphangitis. The result was again negative.

Control tests were made using an anti-yeast serum with the various kinds of yeast used in previous experiments. The anti-yeast serum was obtained from a rabbit which had received a number of injections either subcutaneously or intraperitoneally, of beer yeast. The tests were repeated with the serum of a normal rabbit. In every case there was no haemolysis in the tubes containing the anti-yeast serum and the different antigens, while in the ordinary control tubes and in the tubes containing normal rabbit serum there was haemolysis.

As a final test the serum of an infected animal was tested with cultures of *Leishmania infantum* and *Trypanosoma vespertilionis*, the antigens being cultures of these organisms obtained in Novy-McNeal-Nicolle medium. A similar series of tests was carried out with normal serum. The results indicated that these antigens fail to prove the presence of any sensitiser in serum from infected animals.

In the author's opinion these results appear to support, if not prove, the theory that the causal organism of epizootic lymphangitis is a blastomycete.

The authors believe that infection always takes place through a wound, but they find that it is impossible to transmit the disease with certainty by inoculation of pus. They do not agree with the view that the disease is insect transmitted.

One case is on record of transmission of the disease to the human subject, the infection taking place through a wound.

Treatment.—The authors give details of forty-three cases of the disease treated with "606."

After a few experiments they found that as good results were obtained with the comparatively small dose of 1 gramme as with larger doses. The drug was administered intravenously according to EHRLICH's directions. They have never observed any general disturbance save in one case in which a mule received 3 grammes and showed symptoms of slight colic and diarrhoea.

The effect of the drug is rapid. When the disease is recent and the primary lesion is situated on the middle portion of a limb (about the knee) there is rapid healing, the corded vessels diminish in size and buds that are already appreciable burst and become indurated.

If the disease has been in existence for some time and the initial wound is situated on the lower part of the limb the effect of the injection is in many cases to cause the appearance of fresh buds along the course of the diseased lymphatic vessel. To the inexperienced this might appear to be an aggravation of the

disease, whereas it is in reality a reaction on the part of the animal for the removal of the parasites. Three weeks or a month should be allowed to elapse before the results of the injection are judged.

In the table given, comprising forty-three cases, twenty-seven animals are said to have recovered, while seven are said to be on the road to recovery; the remainder having died or been killed. It is pointed out that in every case in which the animal had to be killed the primary infection occurred in the lower parts of the limbs.

When the diseased lymphatic vessel is well marked and is in a position which renders surgical interference easy this method of treatment should be resorted to, "606" being reserved for those cases that are inoperable.

The case of human infection terminated in complete recovery in three days after the intravenous injection of 0·6 g. of "606."

Prophylaxis.—Before the experiments with "606" were carried out hypodermic inoculations of yeasts were tried, to see whether they might not lead to the production of antibodies capable of exercising an effect on the cryptococci. It was found that there was increasing intolerance of the organism after each inoculation, abscess-formation with discharge of pus occurring within progressively shorter periods. The authors have attempted to make use of this fact in protecting animals in an infected stable by giving half of the animals an inoculation with yeast, leaving the others as controls. Sufficient time has not yet elapsed to enable any statement regarding this experiment to be made.

(170) WALKER (G. K.). *The Treatment of Rinderpest and Haemorrhagic Septicaemia with Permanganate of Potash.*—*N. Comp. Path. & Therap.* 1912. Vol. 25. No. 3. pp. 185-202.

RINDERPEST.

Originally the drug was tried upon two cases of the disease both of which recovered, the dose in each case being $\frac{1}{2}$ dram dissolved in about a gallon of water. Shortly afterwards two further animals were treated, one of these receiving $1\frac{1}{2}$ drams and the other $\frac{1}{2}$ dram. Both of the animals recovered.

Some months later there was an outbreak of the disease in a herd of animals, eighty-eight of which were attacked. Of these 55 had died. Fifteen of the survivors were found to be suffering from the disease, but owing to opposition on the part of the owners it was found impossible to subject all to the treatment. Six were treated, however, the remainder acting as controls.

Subsequently twenty-four additional animals were brought under treatment. Of the thirty animals treated only three died, while of thirteen controls seven died.

The dose of the drug given varied according to the size and age of the animal and the clinical condition. In one or two instances a dose of $\frac{1}{2}$ dram was administered, but as a rule the dose was either 1 or 2 drams. Some of the animals received a single dose only,

while others received as many as four, and in one instance six. The drug was dissolved in about half a gallon of water with an ounce or two of vinegar added.

Post-mortem examinations were made in all the fatal cases and the diagnosis confirmed.

HAEMORRHAGIC SEPTICAEMIA.

Three animals were originally treated, with two recoveries, the third animal being in a critical condition at the time of treatment.

Twenty-three cases in all were treated and recovery followed in ten of these. Since the mortality usually lies between 90 and 100 per cent., the results appeared encouraging.

The percentage of recoveries in cattle was very much higher than in buffaloes (83.3 per cent. as compared with 29.4 per cent.). The greater susceptibility of the buffalo indicates that the larger doses should be tried.

The drug was administered as in the cases of rinderpest. The following is an abstract from the conclusions drawn by the author:—

While no definite conclusions can be drawn from the experiments described they appear to justify the hope that the treatment may prove useful. It is probable that considerably larger doses can be given and with good effect. Further experiments are required to ascertain the maximum dose that can be tolerated.

Calves may be given $\frac{1}{2}$ to 1 dram, medium-sized cattle 2 drams, and animals weighing over 500 lbs. 3 to 4 drams.

It has yet to be decided whether the best results are to be looked for from the administration of a large initial dose or from the daily administration of moderate doses. The former would be the more convenient and in the case of haemorrhagic septicaemia there is rarely time for more than one dose.

The method will have to be tried in cases of rinderpest among animals having little or no natural immunity.

Hypodermic injection of the drug might be tried, but it is probable that its caustic nature would be injurious to the tissues. The injection of the drug direct into the abomasum is worthy of trial.

(171) NICOLAS (C.). *Observation Clinique d'une Affection Chevaline sevrissant à Nindiah et Nindivin (Houailou)*. [Clinical Observation on a Disease of the Horse at Nindiah and Nindivin.]—*Bull. Soc. Path. Exot.* 1912. July. Vol. 5. No. 7. pp. 519-521.

The principal symptom of the disease and the one which first attracts attention is great enlargement of the nasal bones on either side of the median line, producing an appearance resembling that seen in the human subject affected with the disease named "Goundou."

The elevation of the bones on either side of the nose may produce a tumour as large as a fist or larger. Palpation shows that the growth is bony. Upon the surface there is usually a network of dilated veins and the skin covering the enlargement is mobile.

The respiration is impaired and there is a discharge from the nose. The glands in the upper portion of the neck and the glands of the head are enlarged. Neither age nor sex appears to have any influence on the occurrence of the disease.

Later the animal begins to cough, and although the appetite is maintained there is sensible loss of condition. The nasal discharge which was at first mucous becomes muco-purulent and increased in amount. In some cases it becomes sanguinolent.

Further tumour-like growths develop in connection with the shoulders, knees, or hocks, causing great deformation, and in some instances subluxations. Some of the growths on the limbs become converted into abscesses or ulcerate. Death takes place after an interval of some months. Bacteriological investigation is necessary to ascertain the exact nature of the disease.

- (172) LEGER (M.) & BOUILLIEZ (M.). *Sur un Plasmodium des Singes. Passages par Espèces Variées. Action Pathogène.* [A Plasmodium of Monkeys. Transmission to Different Species. Pathogenic Effects.]—*Compt. Rend. Soc. Biol.* 1912. Aug. 2. Vol. 73. No. 28. pp. 310-313.

In the present paper the authors confine themselves to a consideration of the pathogenic effects, and the transmissibility of the parasite to other species, leaving the study of the details of the parasite itself for further study.

The organism was found in one of a lot of five monkeys (*Macacus cynomolgus*) all of which died soon after their arrival at the Institute.

The authors have succeeded in passing the parasite through seven generations in monkeys, and have proved the organism to be pathogenic for the following species: *M. cynomolgus*, *M. sinicus*, *Cynocephalus*, *M. rhesus*, *Cercopithecus patas*, *Cercocebus fuliginosus*. The Chimpanzee, and the Maki of Madagascar have proved resistant.

In some cases death has been very rapid, the monkeys dying in seven days. In other cases the course of the disease is far slower. The authors have some animals which have been infected for more than three months. In such cases relapses are observed.

The authors are of the opinion that the organism resembles the *Plasmodium inui* of HALBERSTAEDTER and PROWAZEK.

- (173) SCHRIDDE (H.). *Die Azur II-Eosin-Färbung an Gefrierschnitten.* [The Staining of Frozen Sections with Azur II-Eosin.]—*Centralbl. f. Allgem. Path. u. Path. Anat.* 1912. July 31. Vol. 23. No. 14. pp. 625-626.

The pieces of tissue should be fixed either in ten per cent. formalin or Formalin-Müller for twenty-four hours, and then washed for a short time in water. The sections are placed in 20 per cent. alcohol for 2 to 5 minutes and then transferred to water. The stain is made up as follows: Azur II-Eosin is added to water, either tap or distilled, in the proportion of two drops to 1 cc. and thoroughly mixed. The sections are placed in this for 25-30 minutes. They are then washed in water for not more than 5 minutes, after which they are placed on slides, carefully dried with filter paper and pressed firmly on to the slides. The slides are then dipped in absolute alcohol eight or ten times, and then

in a second bath of alcohol, and finally in a bath of xylol or toluol about the same number of times.

The sections are then ready for mounting in neutral Canada balsam. The method is said to be of especial value in the demonstration of bacteria and protozoa.

(174) **Extract from a Manuscript Report** (by Mr. R. J. Stordy, Chief Veterinary Officer, East Africa Protectorate) **on a Disease, resembling Mumps, amongst Camels in the Northern Frontier Districts of the Protectorate.**

Reference is made to a peculiar disease affecting camels in the Northern Frontier District. The condition, which appears to be of a contagious nature, somewhat resembles mumps in some of its clinical manifestations. The onset of the disease is rapid. There is no rise of temperature, and the chief, and practically only symptom is an acute and extensive swelling of the glands in the region of the throat. The swelling may be so great as to cause death by asphyxia. Post-mortem examination showed acute inflammation of the larynx and fauces. The glands of the throat were enlarged and oedematous, and the tissues of the throat and the upper part of the neck were infiltrated with a whitish gelatinous material. The report of the results of the examination of material taken from cases is not yet to hand.

Favourable results are said to have followed free opening of the glands, and blistering of the affected parts.

Stordy is of the opinion that the disease is not anthrax.

A request for information as to the occurrence of a similar disease in Egypt or the Sudan elicited the reply that only three cases of disease resembling mumps have been met with in Egypt. In these cases the condition was a benign one occurring among young animals. The symptoms were a disinclination to feed, considerable swelling of the glands of the throat, and some oedema of the face and neck. The swelling was not sufficiently pronounced to affect the respiration, and there was no rise of temperature. Blood examinations were not made. Nothing is known regarding such a disease in the Sudan.

A request for information as to the existence of a similar disease among camels in India elicited the following information. Three cases of an apparently similar disease have been met with. The symptoms were enormous enlargement of the jowl, parotid region and face. Great swelling of the tongue. Two or three degrees rise of temperature. All swellings painful. In one fatal case the duration of the disease was about five days. In one case in which there was partial recovery the illness lasted a week or more.

In the animal which made a partial recovery there was never complete restoration of condition, and the animal suffered from atrophy of the tongue and finally died.

At the post-mortem examination made on an animal four hours after death the following conditions were found. No evidence of anthrax could be found in the blood or in the lesions of the

throat. The spleen was normal in size, the blood was not coagulated. There were extensive haemorrhages in the abomasum, small intestines and tongue; large intestine not inflamed. There was no evidence that the disease was one of the haemorrhagic septicaemias. A brief note is given of a reference to a similar disease occurring in Somaliland by LENNOX-CUNNINGHAM ("Burden Camels," page 15).

BOOK REVIEW.

(175) LAVERAN (A.) & MESNIL (F.). *Trypanosomes et Trypanosomiasés*. 2nd Edition. 1,000 pages, 198 text-figures, and 1 coloured plate. 1912. Paris: Masson et Cie.

Some idea of the enormous strides that have been made in the study of trypanosomes and the diseases for which they are responsible may be gained from the fact that the present edition is more than twice as large as that published in 1904. Reference to the table of contents will show to what extent the various sections of the work have been enlarged, and to what extent fresh material has been incorporated.

The earlier portion of the book which deals with the generalities of the subject has increased from forty-six to two hundred and fifty pages. This increase is largely accounted for by the fact that seven new chapters have been introduced dealing with such subjects as the cyclical and mechanical transmission of trypanosomes, the evolution of the organisms in the invertebrate hosts, the question of reservoirs, cultivation, etc.

In addition to the trypanosomes that are pathogenic for animals and man, the non-pathogenic species found in the former and the trypanosomes of birds and cold-blooded animals receive full attention.

The general plan of the earlier edition is closely adhered to, and there are numerous references which bring the subjects almost up to the date of publication of the book. It may perhaps be noted that the work of observers other than the French has not received as much notice as it might have done. Everyone who is connected with the study of the trypanosomes, whether intimately or not, must have access to the volume, but the value of the book as a book of reference would have been very greatly enhanced had a detailed index been included.

The book is well got up and the type is clear, but it is a matter for regret that some of the illustrations leave something to be desired.

RECENT LITERATURE.*

[Continued from Bulletin No. 1, pp. 53-59.]

Anaplasmosis.

- (176) BEVAN (LI. E. W.). The Anaplasmoses of Cattle.—*Veterinary Jt.*, 1912. July. Vol. 68. No. 445, pp. 392-400.

Babesiasis.

- (177) VON RATZ (S.). Piroplasmosis der Schafe. [Piroplasmosis of the Sheep.]—*Allatorvosi Lapok.* 1912. No. 18; ex. *Berlin. Tierärztl. Wochenschr.*, 1912. July 4. Vol. 28. No. 27, p. 493.
- (178) SEIDELIN (H.). Leishmaniasis and Babesiasis in Yucatan.—*Ann. Trop. Med. & Parasit.*, 1912. July 31. Vol. 6. No. 2, pp. 295-299. With 5 text-figures.

Foot-and-Mouth Disease.

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- (181) MÜLLER (M.). Ueber die Natur der Kugelförmige Gebilde in den Aphthen Maul- und Klauenseuchekranken Tiere. [The Nature of the Rounded Bodies in the Lesions in Animals affected with Foot-and-Mouth Disease.]—*Centralbl. f. Bakt.*, 1. Abt., Orig., 1912. Aug. 24. Vol. 66. No. 1, pp. 103-105.
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- (183) SIEGEL (J.). Bericht über fortgesetzte Versuche mit dem Erreger der Maul- und Klauenseuche. [Further Experiments with the Cause of Foot-and-Mouth Disease.]—*Berlin. Tierärztl. Wochenschr.*, 1912. Vol. 28. No. 39, pp. 713-718.

Leishmaniasis.

- (184) FRANCHINI (G.). On the Presence of Leishmania in the Digestive Tract of *Anopheles maculipennis*.—*Ann. Trop. Med. & Parasit.*, 1912. May 29. Vol. 6. No. 1, B, pp. 41-52. With 3 plates.
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- (185) VISENTINI (A.). Mechanisme de l'Immunité Naturelle du Rat et du Cobaye à l'égard des Cultures de *Leishmania infantum*. [The Mechanism of the Natural Immunity of the Rat and the Guinea-Pig to Cultures of *Leishmania infantum*.]—*Bull. Soc. Path. Exot.*, 1912. June. Vol. 5. No. 6, pp. 358-360.

Malaria.

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- (187) BASS (C. C.) & JOHNS (F. M.). The Cultivation of Malarial Plasmodia (*Plasmodium vivax* and *Plasmodium falciparum*) in vitro.—*Jl. Experimental Med.*, 1912. Oct. 1. Vol. 16. No. 4, pp. 567-579.

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Malaria—continued.

- (188) FERMI (C.) & LUMBAU (S.). Können Anopheles-Mücken auf den Menschen Malaria übertragen, ohne sich durch Besuch von Malariakranken verseucht zu haben? Können dieselben sich die Infektion aus anderen Tieren als dem Menschen holen? [Can Anopheles transmit Malaria to Man without having become Infected from a Previous Diseased Person? Can they become Infected from Animals other than Man?]*—Centralbl. f. Bakt.*, 1. Abt., Orig., 1912. July 3. Vol. 65. Nos. 1/3, pp. 105–112.
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- (202) BLACKLOCK (B.). The Measurements of a Thousand Examples of *Trypanosoma vivax*.—*Ann. Trop. Med. & Parasit.*, 1912. Vol. 5. No. 4, p. 521.
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- (204) BRUCE (Sir D.), HARVEY (D.), HAMERTON (A. E.), DAVEY (J. B.) & Lady BRUCE. The Morphology of the Trypanosome causing Disease in Man in Nyasaland.—*Proc. Roy. Soc.*, 1912. Aug. 24. Vol. 85, No. B 581, pp. 423-433. With 2 plates.
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- (213) MESNIL (F.) & BLANCHARD (M.). Infection comparée des Porcs par *Tryp. gambiense* et *Tryp. rhodesiense*. [The Comparative Infection of Pigs with the *T. gambiense* and the *T. rhodesiense*.]—*Bull. Soc. Path. Exot.*, 1912. July. Vol. 5. No. 7, pp. 492-495.
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- (240) CORTELEZZI (E.). Enfermedad Nodular de los Intestinos. [Intestinal Nodular Disease.]—*Rev. Zootec.*, 1912. Vol. 3. No. 33, p. 696.
- (241) FÜLLEBORN (F.). Zur Morphologie der *Dirofilaria immitis* Leydi 1856. [The Morphology of *Dirofilaria immitis*.]—*Centralbl. f. Bakt.*, Abt. 1., Orig., 1912. July 17. Vol. 65. Nos. 4/5, p. 341.
- (242) HENRY (A.) & BLANC (G.). Le Physaloptère du *Macacus cynomolgus* L. (Nematode).—*Bull. Soc. Path. Exot.*, 1912. June. Vol. 5. No. 6, pp. 390-391.
- (243) LEIFER (R. T.). Check-list of Helminthes Parasitic in Cattle. *Jl. London School Trop. Med.*, 1912. March. Vol. 1. Pt. 2, pp. 115-123.
- (244) NICHOLLS (L.). Windward Isles (St. Lucia) Laboratory Report for Six Months ended March 31st, 1912; (3) Ankylostomiasis in Domesticated Animals.—Report to the Secretary of State for the Colonies.
- (245) RAILLIET (A.), HENRY (A.) & LANGERON (M.). Le genre *Acanthocheilonema* Cobbold et les Filaires péritonéales des Carnivores. [The genus *Acanthocheilonema* (Cobbold) and the peritoneal Filariae of Carnivora.]—*Bull. Soc. Path. Exot.*, 1912. Vol. 5, No. 6, pp. 392-395.
- (246) SCHÜFFNER (W.). Der Wert einiger Vermifuga gegenüber dem Ankylostomum, mit Bemerkungen über die Wurmkrankheit in Niederländisch-Indien. [The Comparative Value of Vermifuges in Ankylostome Infections in the Dutch Indies.]—*Arch. f. Schiffs- u. Tropen. Hyg.*, 1912. Sept. Vol. 16. No. 17, pp. 569-588.
- (247) STEPHENS (J. W. W.). *Paropisthorchis Caninus*. [The Liver-fluke of the Indian Pariah Dog.]—*Ann. Trop. Med. & Parasit.*, 1912. May 29. Vol. 6. No. 1, B, pp. 117-123. With 1 text-figure and 3 plates.
- (248) STILES (C. W.). Third List of Generic Names for the Official List of Zoological Names.—*Parasitology*, 1912. Vol. 5, No. 2, pp. 118-121.

Protozoal Parasites.

- (249) BRUMPT (E.) & JOYEUX (C.). Sur un Infusoire nouveau Parasite du Chimpanze *Troglodytella abressarti*, n. g. n. sp. [A new Infusorial Parasite of the Chimpanzee.]—*Bull. Soc. Path. Exot.*, 1912. July. Vol. 5. No. 7, pp. 499-503. With 1 plate.
- (250) CARDAMATIS (J. P.). De Quelques Microsporidies chez la Mouche Domestique. [Microsporidia of *Musca domestica*.]—*Centralbl. f. Bakt.*, 1. Abt., Orig., 1912. July 3. Vol. 65. Nos. 1/3, pp. 77-79. 1 plate.
- (251) ELLIS (M. M.). Five polycysted Gregarines from Guatemala.—*Zool. Anz.*, 1912. Vol. 39. Nos. 23/24, pp. 680-689. With 7 figures.
- (252) FRANÇA (C.). Sur les Haematozoaires des Taupes. [The Haematozoa of Moles.]—*Arch. Inst. Bact. Camara Pestana*, 1912. Jan. Vol. 3. No. 3, pp. 271-278. With 1 plate.
- (253) HINDLE (E.). What is the Genus *Leptomonas* Kent?—*Parasitology*, 1912. Vol. 5. No. 2, pp. 128-134.

Protozoal Parasites—*continued*.

- (254) MANCEAUX (L.). Hémogregarines du Lézard vert, *Lacerta ocellata* (var. pater.). [The Haemogregarines of the Green Lizard.]—*Bull. Soc. Path. Exot.*, 1912. June. Vol. 5. No. 6, pp. 347-349.
- (255) PLIMMER (H. G.). On the Blood-Parasites found in Animals in the Zoological Gardens during the four Years 1908-1911.—*Proc. Zool. Soc. London*, 1912. Pt. 2, pp. 406-419. With 7 plates.
- (256) PROWAZEK (S. v.). Beiträge zur Kenntnis der Protozoen und Verwandten Organismen von Sumatra (Deli). [Protozoa and Allied Organisms from Sumatra.]—*Arch. f. Protistenkunde*, 1912. July 22. Vol. 26. No. 2, pp. 250-272. With 3 plates and 1 text-figure.
- (257) REICHENOW (E.) & SHELLACK (C.). Streitfragen in der Coccidienforschung. [Controversies regarding Coccidia.]—*Zool. Anz.*, 1912. Vol. 39. Nos. 21/22, pp. 609-617.
- (258) SEIDELIN (H.). Notes on Some Blood Parasites in Man and in Mammals.—*Ann. Trop. Med. & Parasit.*, 1912. Vol. 5. No. 4, pp. 501-507.
- (259) WOODCOCK (H. M.). Notes on Sporozoa. Nos. II, III and IV. No. II. Observations on *Karyolysis lacertae* (Danil.). No. III. Comparison of the Nuclear Condition in Haemogregarines with that of certain Coccidia. No. IV. The Nuclear Structure of Leucocytozoon and Halteridium.—*Quart. Jl. Microscop. Sci.*, 1912. Sept. Vol. 58. No. 1, pp. 171-240. With 2 plates.

Unclassed.

- (260) ARLO. Rapport sur une Épidémie de Péripneumonie sévissant sur la Race Bovine dans la Cercle de Mankono (Côte d'Ivoire). [Épidémie de Péripneumonie of Bovines at Mankono.]—*Ann. d'Hyg. et de Méd. Colon.*, 1912. April-May-June. No. 2, pp. 390-393.
- (261) NYASALAND PROTECTORATE. Annual Report of the Department of Agriculture for Year ended March 31st, 1912. Zomba: Gov. Printers, Nyasaland. Veterinary Division. Report of the Veterinary Officer, pp. 29-49. Trypanosomiasis, Blackwater, Ophthalmia in Cattle, Fowl Cholera, Mombasa Cattle Disease (East Coast Fever), Native Cattle Industry, Dipping, Suspected Rabies, Poisoning among Sheep.
- (262) OSTERTAG. Tierseuchenbekämpfungen in den Colonien, besonders in Deutsch-Südwestafrika. [Campaigns against Animal Diseases in the Colonies, particularly in German South-West Africa.]—*Jahrb. d. Deutschen Lander-Gesellschaft*, 1912. Lfg. 1, pp. 109-116.
- (263) PINOY (E.). Epidermophyton du Singe.—*Bull. Soc. Path. Exot.*, 1912. Feb. Vol. 5. No. 2, pp. 60-63.
- (264) SORRELL (W.) & CASER (F. C.). Rinderpest as Observed in the Philippines.—*Amer. Vet. Rev.*, 1912. Vol. 41, p. 290.

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BABESIASIS.

- (265) MAURITIUS. Bovine Piroplasmosis in Mauritius.—*Annual Report on the Bacteriological Laboratory for the Year 1911*. 1912. Port Louis: Printed at the Government Printing Office. pp. 15-16. With 1 plate.

Piroplasma bigeminum was seen for the first time in Mauritius in a cow that was infected with *P. mutans* and trypanosomiasis. The cow was three years old and owing to the *mutans* infection it was removed to the laboratory for inoculation experiments. While there, a parasite was observed in its blood that presented all the appearances of *P. bigeminum*. While under observation the animal passed blood-stained urine on a number of occasions. A calf was successfully inoculated from this animal.

- (266) SYMONS (T. H.) & PATTON (W. S.). Report on an Outbreak of Canine Piroplasmosis due to *Piroplasma gibsoni* (Patton) among the Hounds of the Madras Hunt, together with some Observations on the Treatment of the Disease with Salvarsan.—*Ann. Trop. Med. & Parasit.* 1912. Oct. 18. Vol. 6. No. 3. B. pp. 361-370. With 5 charts.

Repeated attempts to transmit the disease from jackals to dogs by means of *Haemaphysalis bispinosa* have failed, and opportunity has not yet offered to try transmission experiments with another tick which is fairly common on the jackal and which according to NEUMANN is a new species allied to *Rhipicephalus simus*.

The pack comprised twenty-one couple and when the outbreak was diagnosed 15½ couple were found to be infected. The disease is a very acute one, and among the lesions produced is very great enlargement of the spleen. No haemoglobinuria was observed in any case although there was marked anaemia.

Intramuscular injections of salvarsan in doses of 0.6 gram were given and a single injection sufficed to effect a cure in the great majority of cases. Some of the hounds apparently received the treatment too late, and one or two died from complications.

- (267) NAWROTZKY (N. N.). Zur Piroplasmoseinfektion der Hunde durch die Schleimhaut des Magen-Darmtraktes. [Infection of the Dog with Piroplasmosis by Way of the Mucous Membrane of the Alimentary Tract.]—*Centralbl. f. Bakt.* 1. Abt., Orig. 1912. Oct. 12. Vol. 66. No. 5-6. pp. 417-420.

The fact that a number of observers have suggested or have shown that certain trypanosomes can be transmitted experimentally by way of the intact mucous membrane of the alimentary tract prompted the author to investigate the same question with regard to piroplasms. He states that while it is well known that certain ticks are responsible for the transmission of piroplasms to horses, cattle, and sheep, the method of transmission to the dog is quite unknown, although it has been suggested that *Haemaphysalis leachi* and *Ixodes ricinus* may be responsible.

The author carried out experiments in which the organs of infected dogs were used for the feeding of experimental animals, but fearing that accidental wounds and abrasions in the mouth might afford a means of entrance of the parasite he subsequently restricted his experiments to the administration of blood containing piroplasms, the blood being either given by the mouth, or introduced directly into the stomach by means of a tube.

From a table given it is seen that three full-grown dogs and five puppies were used in the experiments with blood. The blood was diluted with either citrate or salt solution and introduced directly into the stomach. Infection followed in every case. Piroplasms were found in the blood of the puppies on the 2nd, 3rd, 6th, and 7th days after the administration of the blood, and in the case of the dogs on the 5th, 6th, and 7th days respectively. All the puppies and one of the dogs died. It is stated that the symptoms presented by the dogs did not differ from those observed after intraperitoneal inoculation.

- (268) BRANFORD (R.). Trypanblau in the Treatment of Canine Piroplasmosis as occurring in India.—*Veterinary Jl.* 1912. Nov. Vol. 68. No. 449. pp. 643-646.

According to the author canine piroplasmosis is a difficult condition to deal with in India owing to the following facts:

1. There is evidence to show that there are two distinct varieties of piroplasms, one of which is very resistant to trypanblau.
2. Pariah dogs in many cases possess a high if not a complete degree of immunity.
3. The great variety of types of dogs makes it difficult to say whether recovery has been due to treatment or natural resistance.

The dye was injected in 1 per cent. solution in normal salt solution subcutaneously, and the dose varied from 1 cc. to 10 cc. per pound body weight. The author found that within these limits the size of the dose did not appear to be of much importance, for if 1 cc. per pound did not cure 10 cc. would not.

In all, fourteen cases were treated. There were two deaths, eleven recoveries, and one case in which the result was classed as doubtful.

THEILERIASIS.

- (269) WÖLFEL (K.). Ueber den derzeitigen Stand der Impfung gegen das Küstenfieber. [Immunisation against East Coast Fever.] —*Zeitschr. f. Infektionskrankh. Parasit. Krankh. u. Hygiene d. Haust.* 1912. Vol. 12. No. 3. pp. 247-255.

THEILER's successes in transmitting East Coast fever, with the resulting immunity, induced him to attempt to discover some method by which animals might be vaccinated against the disease. He had observed that the disease could only be successfully transmitted by the inoculation of pieces of tissue and consequently directed his efforts to discovering to what extent the reduction in size of the pieces might be carried without vitiating the result. In order to cause embolism in the internal organs, from which he expected to get the same results as from the implantation of larger pieces of tissue, he added peptone or aleuronate to the minced tissues.

By these experiments it was shown that the safest method of inoculation was the intra-jugular injection of a mixture of coarsely minced spleen or lymphatic gland and peptone.

One hundred and thirty-six animals were so treated. Of these ten died of intercurrent diseases, thirty-eight as a result of the inoculation and twenty-one died of the disease when they were being tested as to their immunity.

Experiments were then made on a larger scale.

In infected herds the animals showing clinical symptoms were separated and examinations made for the presence of Koch's granules. If these were discovered the animals were killed and the spleen and peripheral lymphatic glands were removed with aseptic precautions.

These were minced in a machine provided with a disc having perforations about 4 mm. in diameter.

The mince thus obtained was mixed with peptone until the mixture had the consistency of "jam." If on examination it was found that there were no bacteria in the mixture, but that plasma bodies were present in large numbers, the material was used for inoculation, the dose for a full-grown animal being 5 cc. and for a calf 2-4 cc. The inoculations were made into the jugular vein.

Deaths due to embolism of the lungs or heart were very rare.

The resulting reactions varied in intensity and as a rule differed from reactions caused by tick infection.

In the latter case the temperature usually rises about the 13th day and remains elevated for 6-8 days. This is followed by a remission and a second rise, the temperature falling rapidly just before death.

Reactions resulting from the inoculation of either spleen or gland pulp may be divided into typical and atypical reactions.

In the case of typical reactions the temperature curve resembles that given by reactions due to tick-infection. There is a rise on the 13-14th day followed by a remission and a second rise. The first rise of temperature is generally of shorter duration than in the case of tick-infection.

In atypical reactions there is no remission. The febrile attack lasts from 1-4 days, seldom longer. In many cases the temperature curves show several small oscillations at irregular intervals. There are also slight reactions in which the evening temperature remains below 40° C.

In animals that react typically Koch's granules can always be found, while in the atypical cases they are frequently absent.

Death is a frequent sequel to the typical reactions and to those in which the febrile attack persists for more than four days.

In such cases Koch's granules make their appearance in a manner exactly resembling that observed in tick infections. Agamonts are to be found in smears from the spleen or lymphatic glands at or shortly after the onset of fever. Shortly after this dividing forms and gamonts are found. One or more days after the appearance of the granules in the glands *Theileria parva* is to be found in the blood. The number of parasites in the glands, spleen and blood increases up to the time of death.

Koch's granules are generally demonstrable in the atypical reactions. In some cases the granules stain faintly and the nuclei appear vacuolated. Parasites of this kind may be present either alone or together with typical forms and as a rule are demonstrable for a short time only. The author suggests that they may be degeneration forms which are associated with the developing immunity.

If the inoculated animals be subjected to tick infestation after an interval, there is in many cases no reaction or a very brief one during the course of which Koch's granules are not discoverable. In other cases there may be febrile attacks associated with a transitory appearance of Koch's granules. These reactions may also be typical or atypical. It sometimes happens that in spite of the antecedent inoculation parasites appear in the glands (agamonts, dividing forms and gamonts) and *Theileria parva* in the blood.

The variations observed in these reactions must in part be due to variations of the virulence and number of the infective ticks, and in part to the degree of immunity acquired.

The degree of immunity established is not always proportional to the severity of the reaction produced. As a rule a reaction associated with the appearance of Koch's granules confers a high degree of immunity.

On the other hand it has occasionally happened that a weak atypical reaction with the complete absence of granules has been followed by a high degree of immunity.

In practice 343 animals that were certainly not immune have been inoculated. Of these 180 died as a result of the inoculation and 5 died subsequently as a result of natural infection, the remainder (46 per cent.) acquiring immunity.

A further batch of animals numbering about two thousand have also been inoculated and with more satisfactory results, but accurate conclusions cannot be drawn as to the value of the method from this series of inoculations since the inclusion of immune animals cannot with certainty be excluded.

At present the question of the application of the method in healthy districts and to imported animals has not been investigated.

(270) SWAZILAND. Report on the Incidence of East Coast Fever Disease amongst Cattle in Swaziland. [Elder (W. A.), Govern. Veterinary Officer.] (MS. Report dated Oct. 22, 1912.)

The disease first appeared in the Territory in 1902, and in a short time obtained a hold over a large portion of the country. At first a great deal of opposition was encountered on the part of the Swazis, but this was gradually overcome and their co-operation in preventive measures obtained.

In 1909 the following measures for the control of the disease were suggested:—

a. The branding of all cattle in the country, in order to facilitate the tracing of illicit movements of cattle.

b. The formation of concentration camps for infected herds.

c. The slaughter of calves of immune parents in infected areas.

d. The erection of a fence between the infected and the clean portion of the country.

a. Branding was successfully carried out after some opposition, and has proved of value.

b. The formation of concentration camps was not very satisfactory.

c. The slaughter of calves was only carried out in those areas in which immune animals were grazing on ground that was still infected. A period of fifteen months was allowed to elapse between the last death and the declaration that the land was free from infection.

d. A fence was erected running from the Komati River to the Usutu River, a distance of about seventy miles. This fence to a great extent served its purpose of preventing cattle straying from infected to clean ground and *vice versa*.

After branding a grazing ground is appointed for each herd, and thus, in the event of an outbreak, the infected land can be localised.

With the discovery of a reliable dip which could be used at short intervals it was deemed necessary to adopt the dipping of cattle.

The necessary funds having been raised the following measures were undertaken by the Government:—

1. Slaughter of all infected herds, or as many of each herd as was deemed necessary.

2. Full compensation for slaughter or deaths from the disease after control of the herd had been assumed.

3. Isolation of infected ground for two years.

4. Slaughter of all cattle moved without permit without compensation.

5. The building of as many dips as possible in clean and infected country, and the adoption, as far as possible, of compulsory dipping.

The results are given for the first dipping tank erected, and from them the following facts are gathered.

During a period of fifteen months 1,204 cattle were dipped, and the number of deaths occurring at the dip was 311. The percentage of deaths was 25.83 as opposed to the usual percentage of 95 in infected herds. The animals were dipped every fifth day from March to September, and every third day from September to May.

All the herds taken to the tank were infected herds, and were grazing on infected land for the whole fifteen months. The majority of deaths were due to infection prior to the animals being brought to the tank. None of the animals shewed any ill effects from the continued dipping at short intervals. The dip used contained arsenite of soda, soft soap, and paraffin.

There were sixteen tanks in operation at the time of writing and four more were to be added during the year.

By means of the continual dipping, in which it is hoped to include horses, mules, donkeys, goats, and native sheep, a complete eradication of all tick transmitted diseases is looked for.

It is shewn that a great financial saving has been effected by the adoption of the compulsory dipping.

TRYPANOSOMIASIS.

- (271) RODHAIN (J.), PONS (C.), VANDENBRANDEN (J.), & BEQUAERT (J.). *Note sur les Trypanoses Animales du Haut-Katanga.* [Note on the Animal Trypanosomiasis in Upper Katanga.]—*Bull. Soc. Path. Exot.* 1912. Dec. Vol. 5. No. 10. pp. 819-822.

In examining the blood of a number of animals introduced from Rhodesia, including mules, donkeys, oxen, pigs, goats, sheep, and dogs, the authors have encountered *T. brucei*, *congolense*, and *cazalboui*.

In both mules and dogs infected with *T. brucei* (or *pecaudi*) they have found forms in which the nucleus was displaced towards the posterior extremity of the body.

In a puppy inoculated with the blood of one of the mules long, slender parasites with a free flagellum appeared in the blood on the fifth day after inoculation, but when the animal died a fortnight later short thick forms were numerous, and in 4.67 per cent. of these there was posterior displacement of the nucleus. In view of the fact that up to the time of writing no case of human trypanosomiasis had been recorded in Elisabethville, where the examinations were made, the authors feel certain that the trypanosome found was of the *brucei-pecaudi* group, and not the *rhodesiense*.

On the high plateaux in Southern Katanga where only *Glossina morsitans* is found this fly transmits the same animal trypanosomes as in the lower country in the northern part of the Province.

- (272) RODHAIN (J.), PONS (C.), VANDENBRANDEN (J.), & BEQUAERT (J.).—Les Trypanoses Animales au Bas-Katanga et leur Rapport avec les Glossines (3^e Note).—*Trypanosoma denysii* (n. sp.) Parasite de l'Ecureuil Volant. [Animal Trypanosomiasis of Bas-Katanga and their Relationship to the Glossinae (3rd Note).—*Trypanosoma denysii* (n. sp.). A Parasite of the Flying Squirrel.]—*Bull. Soc. Path. Exot.* 1912. Oct. Vol. 5. No. 8. pp. 608-611.

In a previous communication (see this *Bulletin*, Vol. 1, No. 1, p. 40) the authors record the occurrence of trypanosomes of the *cazalboui* and *congolense* types, and the part play by *G. morsitans* in their transmission. In the present note they record the discovery of the *T. brucei* (or *pecaudi*) and give details of experiments in which it was successfully transmitted by *G. morsitans* bred in the laboratory.

A description is added of a new flagellate found in the blood of a flying squirrel.

T. brucei has been found in the blood of a dog, which was at the same time host of *T. congolense*, and also in the blood of a goat.

A guinea-pig inoculated from the dog became the subject of a double infection. Female *morsitans* which were kept in the laboratory for the breeding of pupae were fed on the goat. A large number of the flies became infected and these transmitted the infection to a guinea-pig and two monkeys.

In a single experiment with flies bred from laboratory pupae a positive result was obtained.

Two of the flies from the batch were found on dissection to have a heavy intestinal infection, and in one of them multiplication was commencing in the proboscis. The fly which had successfully transmitted the infection was found to have parasites in the whole of its intestine and in its proboscis.

From the results obtained the authors calculate that 4.16 per cent. of the flies are infective.

Trypanosoma denysii in the fresh state executes active movements which do not involve much translation. One can also make out that the posterior end is drawn out into a point, and that there is a free portion to the flagellum.

In preparations fixed with osmic acid and stained with Laveran-Borrel the trypanosome was found to measure 37-48 microns, and the free portion of the flagellum 8-10 microns.

The width of the body opposite the nucleus is 2-4 microns. Anteriorly the body becomes wider, and posteriorly it is drawn out into a point. The cytoplasm stains uniformly pale blue, and no metachromatic granules or vacuoles are present.

The nucleus is oval and lies in the long axis of the body. It is situated in the anterior portion of the body and does not show a distinct caryosome.

The blepharoplast which is situated about 7 microns from the posterior end of the parasite is large and rounded. The flagellum is thick.

A flying squirrel was under observation for three days during which time the parasites were constantly present in its blood but not in very large numbers.

- (273) ARCHIBALD (R. G.). A Trypanosome of Cattle in the Southern Sudan.—*Jl. Comp. Path. & Therapeut.* 1912. Dec. Vol. 25. No. 4. pp. 292-297. With 1 text-figure.

The trypanosome was originally derived from a cow brought from the Mongalla Province to Khartoum.

In the living state the organism showed very active movements, not associated with any marked translation, and it had a tendency to adhere to the red corpuscles.

The stained preparations were made by fixing moist films with osmic acid and staining either with Leishman or Giemsa.

The parasite measured from 9 to 20 microns in length, and was found to be somewhat short and stout in form. Very squat and tadpole forms were met with, and particularly in films in which the parasites were numerous. The posterior extremity was, in most cases, blunt and rounded. The membrane was fairly well developed, and was as a rule somewhat narrow and straight. There appeared to be no free flagellum, and this structure arose at a point quite close to the micro-nucleus.

The average breadth of the trypanosome, including the membrane, was 1.4 microns.

The majority of the organisms showed a number of chromatin-staining granules posterior to the nucleus, and this was particularly the case in the longer forms. Some, however, showed granules anterior to the nucleus, and vacuoles, when present, were found in the same portion of the body.

The micro-nucleus was terminal or nearly so.

Experiments showed that the trypanosome was pathogenic for the following animals: ox, mule, monkey, goat, sheep, rabbit, donkey, dog, gerbil, and jerboa. The guinea-pig was found to have a relative immunity.

The symptoms and lesions were those usually associated with trypanosome infections. In some animals the trypanosomes steadily increased in numbers up to the time of death. In the goat there was a certain periodicity, and the trypanosomes were usually small and very uniform in length.

No experiments could be undertaken to ascertain the agent concerned in the transmission of the parasite owing to lack of material, but from the history of the original animal the possibility of tsetse acting as the carriers could be excluded. Either *Stomoxys* or *Tabanidae* may have been the transmitting agent.

The trypanosome could be cultivated fairly readily on blood agar. At the end of forty-eight hours numerous clumps of active trypanosomes with their flagellar ends directed outwards could be seen. Stumpy and plasmodial forms which were found after twenty-four hours were less numerous a day later. After seventy-two hours, numerous long forms were present, many of which were vacuolated and contained granules. The trypanosomes lost their vitality after the fifth day.

The author concludes that the trypanosome is the *T. pecorum*.

- (274) BETTENCOURT (A.) & BORGES (I.). Présence de Trypanosomes dans le Sang des Bovidés Portugais. [Trypanosomes in the Blood of Portuguese Cattle.]—*Bull. Soc. Path. Exot.* 1912. Oct. Vol. 5. No. 8. pp. 603-604; & Nov. No. 9. p. 725.

After having examined the blood of eighty animals by the cultural method with negative results, the authors discovered three animals out of a batch of eleven having trypanosomes in their blood. The forms of the trypanosomes as they appeared in the authors' cultures corresponded exactly with those described by other authors, crithidial forms being prominent. The organisms made their appearance in the cultures two days after being sown and persisted for fifteen days or more. In stained preparations, and particularly from cultures of moderate age, the parasites stained deeply and showed large numbers of granules in their cytoplasm. Forms in the process of longitudinal division were encountered in many cases, particularly about the seventh day of growth. Rounded flagellate forms were very rare. Absolutely typical trypanosome forms have been met with somewhat rarely in cultures from seven to eight days old.

In some of these forms the blepharoplast, although posterior in position, was close to or touching the nucleus, but in other specimens the two bodies were at some distance from each other, the blepharoplast being in the posterior third of the parasite, and the nucleus at the junction of the anterior and middle thirds. The parasite possesses an obvious undulating membrane. The authors have not been able to discover parasites enclosed in leucocytes as described by BEHN.

The authors have not been able to find flagellates in the blood of 23 animals from the Azores and they suggest the possibility that the intermediate invertebrate host may not occur in those islands.

- (275) MITZMAIN (M. F.). The Transmission of Surra in the Philippines. [MS. letter.]

In a private communication the author states that he has succeeded in transmitting surra by means of *Tabanus striatus*, the common horse-fly of the Philippines. The experiments were performed with bred flies by the direct method from guinea-pig to monkey and from horse to horse. He states that this fly is undoubtedly the carrier of surra in extensive outbreaks in the Philippines. There exists a decided correlation between the predominance of the fly and outbreaks of surra.

- (276) DUKE (H. L.). A Camel Trypanosome, with some Remarks on the Biometric Method of Diagnosing Trypanosomes.—*Proc. Roy. Soc.* 1912. Oct. 31. Series B. Vol. 85. No. B 583. pp. 563-568.

The trypanosome here described was originally derived from a camel from Boran, and the experiments were undertaken to see whether the trypanosome was transmissible by laboratory-bred *G. palpalis*; a few sub-inoculations were performed.

Morphology.—Measurement of 400 trypanosomes taken at random shewed that the length varied from 18 to 34 microns.

The great majority of the parasites were slender, but a few broad forms were seen. The flagellar [*? aflagellar*] end was in some cases very drawn out, and the kinetonucleus from 4 to 5 microns from this extremity. The undulating membrane was well developed.

The kinetonucleus was well developed but small, rounded in shape, and variable in situation from the extreme end of the parasite up to 4.5 microns from the posterior end.

The nucleus was situated near the middle of the body.

A number of fly transmission experiments were carried out but in no case was a successful transmission obtained, and no trypanosomes were found in the flies used, either in the proboscis or the gut.

Many of the sub-inoculations into other species recorded in the paper were carried out by MONTGOMERY from whom the trypanosome was obtained.

In an ox the disease lasted 112 days, trypanosomes were never observed in the peripheral blood, but infection was proved by sub-inoculation.

In a mule, in one instance the period of incubation was five days. The animal was alive 130 days later, and the trypanosome could not be seen in the blood by direct observation.

In a second mule the only detail given is that the duration of the disease was 136 days.

In two donkeys the period of incubation was eight days, and the duration of the disease 99 and 128 days.

In dogs the period of incubation varied from 7 to 11 days, and the duration of the disease from 21 to 68 days.

In monkeys, rats, and guinea-pigs the period of incubation was four days. The duration of the disease in the rats was 10 days, and in the guinea-pigs about 70.

Identity of the trypanosome.—According to the author the diagnosis appears to rest between *T. brucei*, *evansi*, *equiperdum*, and *equinum*. The presence of a well-marked centrosome and the absence of any plaques excludes the latter two. A curve is given which the author has constructed upon the measurements of 100 parasites from each of four experimental animals, and the curve is said to correspond roughly with BRUCE's curve for *T. evansi*. The absence of short stumpy forms from all the experimental animals is against *T. brucei*.

The animal experiments, although not typical of *T. evansi*, suggest this parasite rather than *T. brucei*, and this conclusion is supported by the evidence furnished by the morphology.

With regard to the biometric method of recognising trypanosomes the author points out that, while in some cases it is of value, its value is in some danger of being over-estimated. To obtain an accurate conception of the dimensions of any trypanosome all stages must be followed out from the commencement to the conclusion, slides taken haphazard being useless. The minute exactness of measurement insisted upon by some authors is held to be irrelevant as regards the practical value of the method.

Other factors which render the biometric method inconstant are :

1. Fixation will vary in different parts of the same slide and in the preparation of slides by different observers.

2. Numerous varieties of strain exist among trypanosomes of any species.

3. The great similarity between many so-called species as regards their length variation.

4. Probably continued maintenance in laboratory animals leads to slight alterations in the morphology of a strain, which to be kept true should be passed from time to time through the insect host.

(277) KINGHORN (A.) & YORKE (W.). Further Observations on the Trypanosomes of Game and Domestic Stock in North Eastern Rhodesia.—*Ann. Trop. Med. & Parasit.* 1912. Dec. 30. Vol. 6. No. 4. pp. 483-493.

This report contains the results obtained from the examination of the game, stock, and small vermin during the whole period spent by the authors in Rhodesia. In an earlier report (see this *Bulletin*, Vol. 1, No. 2, Ref. No. 124) the various trypanosomes found in game and domestic animals in the vicinity of Nawalia in the Luangwa Valley were described. In this abstract the results obtained at Ngoa, to which place the headquarters of the Commission were moved at the end of April 1912, are given. The routine method of examination was that described in the previous report.

At Ngoa, 124 wild animals belonging to 16 genera were examined, and trypanosomes found in 21—a percentage of 16·9. Trypanosomes were found in the peripheral circulation of bucks in 16 instances, but it is probable that had several preparations been made from each buck the percentage of successes would have been higher. In several instances only a single trypanosome was found after prolonged search.

Considering only the animals from which inoculations were made it is found that 23·3 per cent. of the local fauna were affected with trypanosomes. The percentage of big game infected with *T. rhodesiense* at Ngoa was 3·3, while at Nawalia it was 16. Both *T. vivax* and *T. nanum* have been found in game, but no inoculations could be made as susceptible animals were not available for the purpose. Had such animals been available, probably a higher number would have been found to be infected.

It would appear that different species of buck differ widely in their susceptibility. Amongst certain common varieties trypanosomes were never found, or only rarely. Waterbuck, eland, bushbuck, and kudu were the species found to be the most heavily infected.

To a certain extent these differences may be accounted for by the habitats affected by the various species, but specific differences in immunity are probably of much greater importance.

A table is given shewing the species of trypanosomes occurring in each animal in which parasites were found, the information

obtained from the results of inoculation being utilised in the compilation of the table, thus rendering the differentiation of *T. pecorum* and *T. nanum* possible. Double infections were found to be not uncommon. The fact that buck have steadily increased in numbers since rinderpest swept through the country would appear to indicate that their tolerance to trypanosomes is very great.

From the table it may be gathered that among the twelve water-buck examined by either method *T. vivax* alone was found in nine instances, the same trypanosome along with *T. rhodesiense* was found in two cases, and with *T. pecorum* in one. *T. pecorum* was found in four eland and one roan. *T. vivax* occurred in one puku and one duiker, another duiker shewing *T. pecorum* or *nanum*. In a sitatunga *T. ingens*(?) was found.

At Ngoa three goats were examined. *T. nanum* or *pecorum* was found in one, *T. vivax* and *nanum* or *pecorum* in the second, and *T. vivax* and *pecorum* in the third.

The suggestion that small vermin might act as reservoirs for trypanosomes was tested by examining at Nawalia and Ngoa 142 wild rats, 15 wild mice, 1 wild rabbit, 1 giant rat, 1 squirrel, 1 galago, and 2 genet. The results were uniformly negative. It is pointed out that many small vermin in Tropical Africa are nocturnal in their habits and are not, therefore, subject to the bites of *G. morsitans*. Two hundred and fifty-six monkeys (*Cercopithecus pygerythrus*) were examined with negative results.

(278) MAURITIUS. Surra in Mauritius.—*Annual Report on the Bacteriological Laboratory for the Year 1911. 1912.* Port Louis: Printed at the Government Printing Office. pp. 7-14.

The treatment of this disease in bovidae and equidae followed the lines laid down by HOLMES, and in the case of the former gratifying results were obtained. In four herds numbering 181 animals there had been 23 deaths during the three weeks before treatment was commenced, but after the inauguration of the treatment no deaths were recorded. The method of treatment adopted was as follows:—One subcutaneous injection with soamin (2 to 5 g. in 5 per cent. solution) followed by the administration of 10 doses (1 to 3 g.) of arsenious acid in bolus on alternate days.

The results obtained with the equidae were not so good as those obtained by HOLMES, but were encouraging.

It is stated that the success of the arsenic treatment of surra depends upon the strict observation of certain rules which may be summarised as follows:—

The arsenic should be administered in full sub-toxic doses. Should relapses occur the system of dosage should be changed and a dose of soamin administered to clear the circulation before the arsenic is recommended. For animals in advanced stages of the disease treatment should consist of injections of soamin at intervals of three or four days until the animals are in a condition to stand treatment with arsenic. The blood should be examined daily, and after treatment an animal should be kept under observation for two or three months, and a control dog inoculated with blood.

- (279) DODD (S.). *Trypanosoma ingens* in the Mouse Deer (*Tragulus javanicus*).—*Jl. Comp. Path. & Therapeut.* 1912. Dec. Vol. 25. No. 4. pp. 281-285. With 1 plate.

The parasite was discovered in three animals brought from Java to Sydney, two of which died within a few weeks of capture, and the other within eight months. All three were affected with filariasis, but this parasitic invasion was not, in the opinion of the author, sufficient to account for death, and he thinks that the rôle played by the trypanosome may not have been a purely harmless one. There were certain minor differences between the organism encountered in these animals and that described by BRUCE, but MESNIL, who had the opportunity of examining the author's specimens, expressed the opinion that the organisms were identical.

Dodd gives the following measurements:—Average length 93·2 microns, breadth 6·6. The parasite executed somewhat deliberate lashing movements involving distinct but not extensive translation.

Owing to the fact that the animals had been captured only a short time before, it was impossible to make any accurate observations regarding clinical symptoms, and in no case were there any very obvious signs of ill health.

The only abnormality found at the post-mortem examination was a slight general oedematous condition of the tissues.

A few inoculations into rabbits and guinea-pigs failed to cause infection.

- (280) BIOT (R.) & RICHARD (G.). De la Possibilité d'Inoculer le *Trypanosoma lewisi* à d'autres Animaux que les Rats. [The Possibility of inoculating Animals other than Rats with *T. lewisi*.]—*Bull. Soc. Path. Exot.* 1912. Dec. Vol. 5. No. 10. pp. 826-827.

The authors have been able to infect gerboas and dormice with this trypanosome.

In the first gerboa, inoculated intraperitoneally from a rat in whose blood trypanosomes were numerous, trypanosomes appeared on the fifth day, but had disappeared by the thirteenth.

In the second gerboa the period of incubation was twenty days, and the parasites persisted in the blood for seven days.

Rats were also inoculated successfully from the gerboas.

A dormouse inoculated with blood from a gerboa at a time when parasites were present in its blood in a state of division became infected, the parasite appearing in the blood one day later. The trypanosomes multiplied and symptoms of illness made their appearance. The dormouse died on the seventh day, the trypanosomes being scanty at the time of death.

- (281) LAVERAN (A.). Essais d'Immunisation contre des Trypanosomes Pathogènes. [Attempts to immunise Animals against some of the Pathogenic Trypanosomes.]—*Bull. Soc. Path. Exot.* 1912. Dec. Vol. 5. No. 10. pp. 877-882.

In this paper Laveran gives the results of some experiments carried out to repeat those of SCHILLING. SCHILLING's procedure

(see this *Bulletin*. Vol. 1. No. 2. p. 94) was followed and *T. brucei* and *rhodesiense* were used. Laveran found it very difficult to immunise rats against *T. brucei*, as out of twelve rats, each of which received five injections of dead trypanosomes, not one acquired any immunity. With *T. rhodesiense* the experiments were a little more successful, but only a very transient immunity was acquired which was not comparable to that often obtained by an animal when spontaneous recovery from a trypanosomiasis occurs.

Details are given of some experiments in which an attempt was made to immunise animals with sensitised viruses. *T. brucei* and *T. evansi* were used. Laveran concludes that there is little hope that the method will be of value.

- (282) GONDER (R.). Experimentelle Studien mit Trypanosomen und Spirochaeten (Spirochaeten). [Experiments with Trypanosomes and Spirochaetes.]—*Zeitschr. f. Immunitätsforsch. u. Exp. Therap.* 1. Teil., Orig. 1912. Nov. 22. Vol. 15. No. 2-3. pp. 257-292. With 1 plate.

This article gives the details of a considerable number of experiments showing the effects of certain staining compounds on ordinary trypanosomes and on arsenic-fast trypanosomes *in vitro*. In the second portion, the details are given of some experiments showing the effects of the serum of animals treated with compounds of arsenic upon certain spirochaetes.

The author's conclusions are as follows:—

1. It can be shewn that while normal trypanosomes can be stained while still alive with certain substances, arsenic-fast organisms are only stained after they are dead.
2. Certain substances cause the blepharoplast of many trypanosomes to disappear. The substances act directly on the blepharoplast, which in the case of the *T. lewisi* tends to approach the nucleus. The auto-oxidation of LAVERAN and ROUSKY does not appear to take place.
3. Trypaflavin, Arsenophenylglycin, and Salvarsan maintain their fixation in the trypanosomes *in vitro*. Mice cannot be infected with trypanosomes treated with these chemical substances.
4. The serum of an animal injected with salvarsan is active against *S. recurrentis* and *S. gallinarum*, *in vitro*, shortly after the injection has been given, and is of therapeutic value.

- (283) THOMSON (J. G.) & SINTON (J. A.). The Morphology of *Trypanosoma gambiense* and *Trypanosoma rhodesiense* in Cultures: and a Comparison with the Developmental Forms described in *Glossina palpalis*.—*Ann. Trop. Med. & Parasit.* 1912. Oct. 18. Vol. 6. No. 3. B. pp. 331-356. With 3 plates.

The majority of the authors' cultures were made with a modified NNN medium, the composition of which was as follows: Agar, 14 g. previously soaked in constant changes of distilled water for thirty-six hours; sodium chloride (pure), 6 g.; distilled water 900 cc. Citrated rats' blood (2 of blood and 1 of 1 per cent. citrate) was added to the melted agar at 45° C to make a mixture of equal parts. The tubes were heated to 45° C for half an hour to destroy the complement and were then sloped. The tubes were

incubated for two days at 25° C to allow of the expression of the water of condensation. There was a considerable degree of uncertainty in all the media tried and a number of tubes had to be inoculated in every case. In inoculating tubes only three or four drops of infected blood should be added to each cubic centimetre of condensation liquid. The optimum temperature for obtaining good cultures which did not die quickly was 22° to 24° C.

The sixth or seventh day was usually most suitable for obtaining subcultures.

It was difficult to obtain good stained preparations from the cultures owing to the salt and free haemoglobin, but it was found that the best preparations were obtained by adopting the following procedure: A drop of the culture liquid is placed on a slide and exposed to the vapour of 4 per cent. osmic acid for thirty seconds, after which it is spread with cigarette paper, again exposed to the osmic acid vapour for a few seconds, and then fixed with methyl alcohol for twenty minutes. The preparations were then stained with Giemsa.

T. gambiense was cultivated for thirty-seven days, during which time it was carried through four generations. *T. rhodesiense* lived only twenty-one days, all flagellates disappearing in the third subcultures.

The forms found in the culture tubes were identical in their chief characteristics with those described by BRUCE as occurring in the gut of an infected fly.

The cultures become non-infective as soon as blood-forms disappear; they were non-infective after the third day.

The authors suggest that the failure of cultures to transmit the infection after this period is due to the fact that in cultures the infective form of the parasite, that found in the salivary gland of the fly, does not develop, a special environment being required.

The authors were unable to find any evidence of a sexual cycle.

(284) ROUDSKY (D.). Sur un Corpuscule Temporaire de *Trypanosoma lewisi* et de *Trypanosoma duttoni*, simulant, à certaines Phases de son Evolution, un Deuxième Noyau. [A Temporary Body in *T. lewisi* and *T. duttoni* simulating in some of its Phases a Second Nucleus.]—*Compt. Rend. Soc. Biol.* 1912. Dec. 27. Vol. 73. No. 37. pp. 730-732. With text-figures.

In stained preparations of the blood of the mouse and rat, containing respectively *T. lewisi* and *T. duttoni*, some of the parasites were observed to contain a body which in certain stages of its development resembled a nucleus. The body was found close to and just in front of the centrosome. It was distinguished from the nucleus by its staining more faintly, and by its rounded form.

The structure in question was about three or four times the size of the centrosome, and was at first filled with small basophile granules. As the body increased in size the granules became aggregated and presented an appearance suggesting a large caryosome. Subsequently the body became still larger and the granules became disposed for the most part at its periphery. Later these disappeared, and the body became converted into a large

vacuole staining a faint pink colour. At this stage the structure was larger than the nucleus and caused bulging of some of the parasites.

In one instance the author observed in the situation occupied by this body a homogeneous mass stained blue like the cytoplasm.

The trypanosomes possessing this body practically always had a very long posterior portion.

The structure has not been observed in dividing forms, nor has the author observed it in *T. lewisi* in the rat and *T. duttoni* in the mouse.

- (285) ROBERTSON (Muriel). *Notes on the Life-History of Trypanosoma gambiense, etc.* [Abstract.] — *Proc. Roy. Soc.* 1912. Dec. 17. Series B. Vol. 86. No. B 584. pp. 66-71. With 27 text-figures.

I. Endogenous cycle in the blood.

The short form may be looked upon as the adult blood-type. It is present almost exclusively during the periods when the parasite is scanty in the blood. When multiplication is going on intermediate, long slender, and dividing forms occur. The short forms appear to be responsible for carrying on the infection in the *Glossina*, and the blood of a monkey is only infective when they are present in sufficient numbers. Intracellular multiplicative forms do not occur in the lungs, liver, or spleen in monkeys.

Round non-flagellate forms are occasionally found in the liver and lungs, apparently between and possibly within cells. They appear at a time when the trypanosomes are being destroyed, but have only been found in a teeming infection examined during the earlier months of the disease. They are apparently about to be destroyed, but their survival in very small numbers as latent forms cannot be excluded.

The differentiation into long and short forms is not an expression of sex.

II. Exogenous cycle in the fly.

While the series of changes undergone in the *Glossina* up to the time of becoming infective is very definite and constant, the duration of the cycle varies within the limits of more than a fortnight. Trypanosomes never attach themselves while in the gut, nor do they ever disappear from this situation at any period; the development occurs free in the lumen from the start. The parasite enters neither the body cavity nor the body cells.

The earliest processes are characterised by slight and indefinite changes of form. Broad, slender, and degenerating specimens are all present, but only the broader types are found in division at this early stage. These divisions show a suppressed crithidial phase in the young individual. This disappears before the separation of the two products. This peculiarity has never been observed after the 10th day. No other crithidial phase is shown in the gut. The parasites generally start developing in the middle or posterior intestine, and by the 7th to 10th day a large number of trypanosomes are present. The blepharoplast plays the role of a centrosome in the division of the kinetonucleus. The division is not

longitudinal, but practically transverse. Division is often unequal.

Very slender forms are developed about the 8th to 18th day and gradually pass forwards into the proventriculus. This form is the culmination of the development in the gut. The parasites may overflow into the sucking stomach, but are not permanently established there, and they are unable to retain their position if the fly be starved for two or three days.

Up to the 10th to 15th day multiple forms containing a number of nuclei may be seen in certain cases. The evidence is largely in favour of these being degenerative forms, but it is not sufficient to exclude the possibility that some of them may be involution or resting forms. Proventricular forms when injected into clean monkeys do not cause infection.

The long slender forms come forward into the hypopharynx in small numbers at a time. From the hypopharynx they pass backwards along the ducts of the salivary glands. They attach themselves where the narrow duct joins the slightly broader part which leads to the glandular portion proper. They become shorter and broader, and assume a crithidial form. Multiplication occurs and gradually the whole gland is invaded. They develop into trypanosomes almost identical with the blood-forms, but are often a little shorter.

There is the strongest presumptive evidence for considering that these types produce the infection in the vertebrate. It is considered that the development in the glands is the essential part of the cycle, the development in the gut being a somewhat indifferent multiplication to enable the trypanosomes to reach the salivary gland, which alone appear to be able to stimulate the trypanosomes to the apparently essential reversion to the crithidial type.

Sexual differentiation has not been observed; this, however, is not a characteristic feature of flagellate life. Isogamy appears to be usual. The evidence of conjugation is slight, but general theoretical considerations are strongly in favour of the occurrence of some such process.

(286) KINGHORN (A.) & YORKE (W.). On the Influence of Meteorological Conditions on the Development of *Trypanosoma rhodesiense* in *Glossina morsitans*—*Ann. Trop. Med. & Parasit.* 1912. Oct. 18. Vol. 6. No. 3. B. pp. 405-413.

In a previous report attention has been drawn to facts which indicate that meteorological conditions have considerable influence on the development of the trypanosome in the fly, and more recently valuable evidence has been obtained indicating that such conditions, and particularly temperature, have a very pronounced influence in this respect.

The present paper is a synopsis of the experiments that have been carried out at Nawalia and Ngoa, the former having been already published.

In the Nawalia experiments 330 flies were used, six, and probably ten, of these becoming infective. In five experiments on the plateau 520 clean flies, 'wild' and 'bred,' were used without a single one becoming infective. The most striking difference in

the climatic conditions met with in the two places was the temperature, there being a difference of 15° to 20° F., that at Nawalia being the higher. In order to ascertain the influence of the temperature on the development of the trypanosome in the fly, a further series of experiments was made on the plateau in which an incubator was used, the flies by this means being kept at a temperature approximating to that encountered in the valley at the most favourable season.

No water was placed in the incubator and the warm dry air had a very deleterious effect on the insects, for within a week of the commencement of the incubation 25 out of 61 in the first batch, and 53 out of 72 in the second had died off. Nevertheless two of the first batch and one in the second proved to be infective. It seems obvious therefore that a high temperature favours the development of the trypanosome in the fly, and this would account for the failures in the experiments carried out at the temperature of the laboratory. From an analysis of the positive experiments carried out in the valley it is seen that the majority of successes were obtained in the hot season.

An occasional wild fly was found to be infective on the plateau, although these results were not obtained in the laboratory experiments, and the explanation probably is that the few infected flies found had been infected during the warmer season of the year and had survived to the cold season.

Comparing the results obtained by feeding freshly caught flies on healthy monkeys in the valley with those on the plateau it was found that in the former place one fly in 534 was infective, while in the latter the proportion was 1 in 1,260. The general circumstances would appear to indicate that the only essential difference between the two places was a difference of climatic conditions. As an additional reason for holding this view it is pointed out that in the valley a greater proportion of infective flies was caught in the hot weather than in the cold season.

An additional factor in the development of the trypanosome in the fly possibly is the relative humidity of the atmosphere, but no definite information has been obtained on this point.

In a further experiment it was found that appearances suggest that the first part of the cycle of development can take place at comparatively low temperatures whereas a considerably higher temperature is necessary for the completion of the cycle. It would appear that the first portion can be effected at 60° F. while for the completion a temperature of $75-85^{\circ}$ is necessary.

(287) KINGHORN (A.), YORKE (W.), & LLOYD (LL.). On the Development of *Trypanosoma rhodesiense* in *Glossina morsitans*.—*Ann. Trop. Med. & Parasit.* 1912. Dec. 30. Vol. 6. No. 4. pp. 495-503.

The authors found that in every fly capable of infecting animals the salivary glands were invaded, and that in flies that were not capable of infecting animals these glands were not invaded by the parasite, although about 20 per cent. of them had trypanosomes in their intestine. A precisely similar state of affairs was

observed in the dissection of 'wild' *G. morsitans*. Of the laboratory bred flies, five out of 132 were capable of transmitting infection, and the salivary glands of 14 out of 620 wild flies were found to be invaded by the trypanosome. All but four of these were definitely proved to be capable of transmitting the trypanosome, the animal upon which the four fed dying before a diagnosis could be made. None of the flies in which the salivary glands were not involved were able to transmit the trypanosome. Similar results were obtained when wild flies were fed on healthy monkeys and the infecting fly isolated.

In order to anticipate the criticism that the parasites were not really in the glands but outside these structures as a result of contamination with the gut, the dissections and examinations were conducted with extreme care. Trypanosomes were found in such numbers as to exclude any possibility of their being outside the glands, and sections of the glands shewed them to be packed full of parasites.

Trypanosomes were present in the intestines of many flies that were examined within a few days of an infected meal, but most of those dissected after the first five or six days were negative. In a certain proportion multiplication of the parasites occurred in the intestine.

The authors have little information as to the reason for this multiplication in the gut of occasional flies only. In the mid-gut of one fly, which died on the twelfth day after having been fed on a guinea-pig infected with *T. rhodesiense*, cysts ranging from 27 to 32 microns in diameter were found. Some of these cysts had thin walls and were filled with a seething mass of flagellates, others had thicker walls and their contents were quiescent. The authors are unable to say whether the fly was infective at the time of death. An animal on which it had fed three days before death did not become infected, and a monkey inoculated with its intestinal content died from an unknown cause two days later.

It was found that inoculation with gut contents failed unless there were also trypanosomes in the salivary glands, and it appeared that if trypanosomes were present in both situations material from these parts was capable of setting up infection on inoculation.

It is uncertain how the salivary glands become infected, but there is a certain amount of evidence to shew that it is secondary, and that it only occurs when the trypanosomes in the gut have reached a certain stage of development, and only then when the conditions of temperature are suitable for the further development of the parasites. Although parasites can develop and multiply in the intestines at fairly low temperatures, trypanosomes were never found in the salivary glands of flies that had not been subjected to temperatures of 75-80° F. Three flies that had been kept at the temperature of the laboratory for forty days after an infecting feed became infective in eight days or less when incubated at 85° F.

The invasion of the salivary gland was observed in flies infected with *T. rhodesiense* only, and never with any of the other trypanosomes dealt with either in the Luangwa Valley or on the Congo-Zambesi watershed.

The authors believe that the presence of trypanosomes in the proboscis of flies is of no special significance, but that it depends upon the passage of infected salivary secretion, or upon regurgitation from the gut during handling.

It is of interest to note that of the 310 'wild' *G. morsitans* dissected as they were brought to the laboratory, recognisable mammalian red corpuscles were found in the intestine of 70, whilst nucleated red corpuscles were found on four occasions only.

Morphology of the trypanosome in *G. morsitans*.—

In a short paragraph it is stated that the form of the parasite occurring in the salivary glands approximates to the short variety in the mammalian blood, but is not identical with it. The predominant type in the intestine is a large broad flagellate, with a feebly developed undulating membrane, and little, if any, free flagellum. The nucleus is fairly central in position, but not infrequently lies a little behind the central point.

A full description of the parasite as it occurs in the fly is left for a further communication.

(288) HALBERSTAEDTER (L.). *Versuche mit einem spontan arsenfesten Trypanosomenstamm*. [Experiments with a Strain of Trypanosomes naturally Resistant to Arsenic.]—*Arch. f. Schiffs- u. Trop.-Hyg.* 1912. Oct. Vol. 16. No. 19. pp. 641-647.

The author points out that the great bulk of the literature regarding the occurrence of drug-resistant strains of trypanosomes refers to strains that have been rendered resistant by experiment.

He states however that, apart from such experimentally produced resistance, different species of trypanosomes are variously affected by certain active chemical substances. Trypanrot is effective against *mal de caderas*, but is practically without effect in nagana. Nearly allied species of spirochates and malarial parasites shew varying degrees of susceptibility to drugs: atoxyl is effective against *S. gallinarum*, but not against *S. duttoni*; salvarsan is more valuable in the case of tertian malaria than in quartan.

Great variation has been observed in the resistance offered by different strains of the same species of trypanosome, but up to the present no record has been made of so high degree of resistance as to be almost absolute against such active arsenical compounds as arsacatin, salvarsan, etc.

The strain in which the author found natural resistance to arsenic was a *mal de caderas* strain originally obtained from Brazil, which had, since its arrival in Europe, been passed through about 720 passages in mice and rats. Neither the original horse, nor any of the animals through which the strain was passed had been treated with arsenic. The discovery that the strain was resistant to atoxyl was made accidentally.

In the experiments which were subsequently carried out, it was found in the first place that the strain was absolutely resistant to arsacatin. The maximum safe dose for a mouse, viz., 1 cc. of a 4 per cent. solution per 20 g. body weight, had not

the slightest effect on the course of the disease. The strain behaved in a manner exactly similar to a strain that had been rendered resistant experimentally. The resistance of the strain to arsenophenylglycin and salvarsan was then tested.

Maximum safe doses of salvarsan were unable to alter the course of infection in the least in experimental animals. In the absence of another strain of the trypanosome strictly comparative tests could not be made, but controls of a kind were furnished by a strain of nagana. Neither salvarsan nor arsacetin in the dose mentioned was found to have any effect on the caderas strain, while both effected cures in the animals infected with nagana.

Arsenophenylglycin (1:1000) delayed the course of the infection slightly, but a single injection of 1 cc. per 20 grammes of a 1:600 solution effected a permanent cure. With the nagana strain 1:1000 solution did not always cause a complete disappearance of the trypanosomes, and 1:1500 produced only a temporary recovery. The nagana strain behaved like an ordinary strain to substances other than arsenic compounds, such as parafuchsin.

The mal de caderas strain was easily affected by potassium antimony tartrate, and to this drug behaved exactly like the normal nagana strain.

Since atoxyl and arsacetin are inactive against trypanosomes in vitro, while salvarsan is active, test-tube experiments were made with the latter. Equal quantities of blood containing trypanosomes and salvarsan solution were mixed together, and the mixture inoculated intraperitoneally into mice after a contact of 20 minutes. The trypanosomes were examined microscopically from time to time.

With the normal nagana strain it was found that salvarsan in 1:100000 solution rendered the trypanosomes unable to infect an animal in 20 minutes, and that the power of infecting an animal was only retained by the trypanosomes when the solution used was 1:200000. With the caderas strain, on the other hand, a 1:2000 solution was required to produce the same effect.

(289) DUKE (H. L.). Some Experiments with Arsenphenylglycin and *Trypanosoma gambiense* in *Glossina palpalis*.—*Proc. Roy. Soc.* 1912. Dec. 17. Series B. Vol. 86. No. B 584. pp. 19-31.

In the experiments detailed in this paper attempts were made to obtain answers to the following questions:—

1. Does the presence of arsenic in the blood ingested by a positive fly destroy the trypanosomes in that fly?

2. Does preliminary feeding of flies on blood containing arsenic have any effect on the subsequent development of trypanosomes in their interior?

3. If flies are fed on blood containing arsenic shortly after the infecting feeds on a gambiense monkey, are the flagellates still capable of development in that fly? If they can still develop is the resultant strain arsenic-resistant in the blood?

4. Has arsenphenylglycin any prophylactic action against the bite of a fly infected with *T. gambiense*, and, if so, what is the extent of this protection.

The experiments devised to furnish an answer to the first of these questions yielded results from which the following conclusions were drawn:

Flagellates in the gut of flies fed upon a monkey within 24 to 48 hours of the administration of arsenphenylglycin in doses of 0.1 g. per kilogramme are markedly affected. The flagellates in the salivary glands are not injured in any way, and the fly does not lose the power of infecting.

In the first pair of experiments devised to answer the second question no positive flies were found either among the arsenic or the control batch.

In a second experiment in which a single batch of flies was used for feeding on a monkey having arsenic in its blood before being placed on an infected monkey, it was found that 2.6 per cent. contained flagellates, while in a control batch the percentage was 11.5.

It is pointed out that care must be exercised in drawing any conclusion from the limited evidence furnished by this experiment. From a small number of experiments carried out with the object of answering the third question it appeared that ingestion of arsenic blood immediately after infecting feeds checks subsequent development of flagellates in the fly.

No experiments appear to have been made to furnish an answer to the second part of the question.

With regard to the experiments devised for supplying an answer to the fourth question the following points must be taken into consideration as having important bearing upon the subject:

a. The mere introduction of the proboscis of a positive fly into the skin of its victim can produce infection. A fly need not withdraw blood to infect an animal. This supports the view that the flagellates in the salivary gland are responsible for infection.

b. A single positive fly can infect a monkey on three consecutive days.

c. The facts recorded in Section 1 relating to the persistence of the salivary gland flagellates and the infecting power of the fly even after the gut has been cleared by arsenic feeding. It was found from this series of experiments that arsenphenylglycin in doses of 0.1 g. subcutaneously per kilogramme protected a monkey against infection by positive *G. palpalis* if given within 12 days before exposure.

Half this dose per kilogramme protected a monkey against infection if given within seven days of exposure.

(290) MASON (E.). **Experimental Treatment of Camels for Trypanosomiasis.**—Dept. of Public Health (Veterinary Section), Cairo. Paper No. 5. 1912. pp. 3-4.

Reference is first made to an earlier series of experiments in which atoxyl, orpiment, and sodium arsenate were used, and which were recorded in detail in the *Journal of Comparative Pathology and Therapeutics*, June 1911.

The present series of experiments was carried out with "606."

It was found by experiment that the requisite dose was approximately 0.0187 gramme per kilogramme body weight, i.e., about

7 grammes for a camel weighing 375 kilogrammes. The drug was administered intravenously.

A batch of eleven camels was placed under observation, the blood being examined whenever the temperature rose above normal. In the case of nine of these animals the trypanosomiasis was complicated with filariasis. The injections of salvarsan were only given when treatment became urgent, the remaining camels acting as controls until they in their turn required treatment.

In some cases the worm embryos ceased to appear quite independently of the injections. Nine of the animals were under observation for periods varying from four and a half to ten and a half months after treatment without the return of the trypanosomes being recorded. White rats inoculated with blood from these recovered animals failed to react in every case.

In the remaining two camels trypanosomes reappeared in the blood in one and three months respectively. In these cases the dose of "606" was not sufficiently large, only six grammes being administered, while the camels weighed 404 and 384 kilogrammes respectively.

During the treatment temporary blindness was observed in four cases. Attempts to obviate this risk by administering smaller doses failed to produce dispersal of the trypanosomes, though there was no blindness. It seems necessary to give a single dose sufficiently large to disinfect the whole system, or the trypanosomes are not killed. The survivors appear to develop a tolerance to the drug, and a second dose would have to be greatly increased. This would defeat the object of dividing the treatment into two doses, and render the first dose unnecessary.

The camels do not require more than the ordinary amount of rest for recuperation—one or two months.

(291) DE GREEF (M.). Guérison de Deux Cas de Trypanosomiase du Cheval par l'Orpiment. [Recovery of Two Cases of Equine Trypanosomiasis under Treatment with Orpiment.]-*Ann. de Méd. Vét.* 1912. Oct. Vol. 61. No. 10. pp. 546-550.

The two cases occurred while the stud at Yakoma was being transferred to Vankerkovenille (Congo) and the parasite responsible was identified as *T. cazalbori*.

The first case was observed in an eight-year-old mare. The symptoms presented were at first very vague but became progressively more marked. There was weakness, emaciation, and later oedema of the limbs, but no very marked rise of temperature. In one eye there was conjunctivitis followed by keratitis and ulceration. The glands of the throat became greatly enlarged, but were not painful on pressure. Trypanosomes were found in small numbers in the blood, but liquid obtained by puncture of the enlarged glands proved negative. Orpiment and tartar emetic were administered, the former as an electuary with extract of opium, and the latter, dissolved in salt solution, was injected into the jugular. The animal received three doses of orpiment of eight and six grammes, and two injections of emetic of one and a half grammes each, within nine days. The trypanosomes disappeared

from the circulation and the animal so improved in condition as to be able to travel three weeks after the commencement of the treatment.

The second animal was only found to be infected with trypanosomes as a result of microscopic examination of the blood, to which all the horses were subjected after the discovery of the first case. The only symptom presented by this case was unilateral keratitis which responded to treatment.

In the first instance this animal received treatment with emetic only as no orpiment was available. Four injections were given, but trypanosomes were still present in the blood nearly three months later. Three doses of orpiment were then given at intervals of one day, the doses being 3.5 to 4 grammes. Trypanosomes were not discoverable in the blood after this treatment.

(292) HOLMES (J. D. E.). *Some Experiments in the Treatment of Surra in Camels.*—*Indian Civil Veterinary Dept. Memoirs.* No. 3. [Period covered 1910-1911.] pp. 78-97.

In the experiments detailed in this paper arsenious acid, arsenic tri-sulphide, atoxyl, soamin, tartar emetic, and lithium antimony tartrate were tried alone and in combination.

In the first instance, the effect of a single dose of varying amount was observed, in order to ascertain the amount of the drug which could be tolerated, and the effect of the drug on the trypanosomes in the circulation.

As the periods of intermission in Surra among camels are very irregular, the interval during which trypanosomes are absent from the circulation after a dose of arsenic or other drug gives only a relative indication of the effect of the dose.

The camels used in the experiments had contracted the disease naturally and were in a fairly advanced stage. Some of the animals died from exposure and from dietetic troubles.

It was found that single doses of arsenic up to ten grammes were ineffective in producing a permanent dispersal of the trypanosomes, and that toxic symptoms were produced by quantities of about five grammes and upwards. Similar results were obtained when larger quantities of arsenic were given in a number of doses at intervals of some days.

A dose of one hundred grammes of tartar emetic was found to produce toxic symptoms without proving effectual in dispersing the trypanosomes. Quantities of five hundred grammes and over, administered in smaller doses, and at intervals of a few days over a period of about a month were found to be ineffectual.

The results obtained with arsenic combined with tartar emetic were better in that in some cases trypanosomes were absent from the circulation for some time after the cessation of treatment, but in the two instances in which this happened the camels died.

Treatment with arsenic in conjunction with atoxyl either proved ineffectual or fatal.

Treatment with ten grammes of arsenic and twenty grammes of soamin also proved fatal.

A single dose of ten grammes of lithium antimony tartrate proved ineffectual, as did a single dose of ten grammes of atoxyl.

Arsenic trisulphide proved fatal in a dose of twenty grammes administered in a drench. Soamin in a dose of ten grammes subcutaneously also proved fatal.

Trypanblue injected subcutaneously in a dose of a little less than two grammes was without effect, and the skin at the seat of injection sloughed.

(293) LANFRANCHI (A.). Sur le Diagnostic des Trypanosomiasés. Essais d'Identification des Différents Trypanosomes. [The Diagnosis of the Trypanosomiasés. Attempts to Identify Various Trypanosomes.]—*Bull. Soc. Path. Exot.* 1912. Oct. Vol. 5. No. 8. pp. 611-614.

Referring to the publications of numerous authors who have done work in connection with the detection of specific antibodies in the serum of animals affected with trypanosomes, either for purposes of diagnosis, or for the differentiation of different parasites, the author points out that their results are not in concordance. He believes that the divergences of opinion may be accounted for by the different antigens sought, and by differences in the course of the infections, for while in chronic diseases antigen formation is abundant, the opposite is the case in the acute or subacute conditions.

Exact details of the procedure are not given, but the author states that, thanks to a special method, he has been able to immunise dogs against nagana, and experiments have been made with the sera of these animals in conjunction with the sera of rats, guinea-pigs and dogs infected with *T. brucei* and *T. equiperdum*.

The serum of the immunised dogs agglutinated the corresponding trypanosome in dilutions of 1 in 75,000, but he was unable to identify the *T. brucei* by means of the serum because it caused agglutination of *T. equiperdum* in dilutions of 1 in 50,000 to 1 in 60,000.

Precipitin reaction.—The author found that when the serum of a hyperimmunised dog was placed in contact with the serum of animals infected with nagana in the proportion of one to three clear results were obtained.

Negative results were obtained with the hyperimmune serum when used with serum derived from an animal infected with *T. equiperdum*.

If however the blood was obtained when trypanosomes were very numerous a positive reaction could be obtained, especially when the experiments were made with guinea-pig serum.

Positive reactions were obtained at once in almost every case. At first there was a slight haziness which within an hour or so became flocculent, and in tubes incubated at 37° C. for four hours fell to the bottom. Sera heated to 55° C. for fifty minutes gave still more obvious reactions.

Fixation of the complement.—The author has found that if a clear reaction is obtained when the blood used is rich in trypanosomes, the effect is even more distinct when the blood is taken during a trypanolytic crisis.

Positive results were also obtained when the hyperimmune serum of the dog was used with the serum of animals infected with *T. equiperdum*.

Conclusions:—

1. Trypanosomes cannot be identified by the agglutination test with hyperimmune sera.
2. Under certain circumstances the trypanosomes of nagana and dourine can be distinguished by the precipitin test.
3. The complement test can under certain conditions be used for the diagnosis of trypanosomiasis, but differentiation of trypanosomes cannot be effected by its means.
4. The use of a hyperimmune serum has enabled the author to produce evidence of the presence in the blood of affected animals of considerable quantities of antigens.

(294) KNUTH (P.) & BONGER (C.). *Nachweis von Trypanosomen bei einem Schlachtochsen mit Milzschwellung*. [The Demonstration of Trypanosomes in an Ox showing Enlargement of the Spleen.]—*Berlin. Tierärztl. Wochenschr.* 1912. Oct. 31. Vol. 28. No. 44. p. 804.

The animal appeared to be in perfect health, but when slaughtered was found to have a greatly enlarged spleen. On a previous occasion examination of specimens from a similar case proved negative, but in this instance trypanosomes closely resembling those first described by BEHN in 1910 were found in smears from the liver, spleen, and kidneys. The trypanosomes varied from 50 to 70 microns in length, and from 4 to 6 in breadth. One specimen was found measuring 99 microns. In preparations stained by Giemsa the cytoplasm was found to contain a large number of reddish granules of various sizes. The parasites were very scanty.

(295) BEHN. *Weitere Trypanosomenbefunde beim Schafe*. [Trypanosomes in Sheep.]—*Berlin Tierärztl. Wochenschr.* 1912. Dec. 12. Vol. 28. No. 50. p. 934.

In October 1911 the author recorded the occurrence of trypanosomes in a sheep, and in the present short note he records their presence in three animals. Two of the sheep were purchased out of the same flock as that from which the previous animal was obtained, and the third was a six-months-old lamb born of one of these animals. The trypanosomes were exceedingly scanty, and could only be demonstrated in thick films stained by Giemsa. One of the striking characters of the trypanosomes was the large size of the blepharoplast. He was unable to cultivate them in blood-broth.

Seven other sheep obtained from a different source shewed no trypanosomes. The author is unable to say whether any connection exists between the *Crithidia melophagia* and the trypanosome of the sheep.

- (296) RODHAIN (J.), PONS (C.), VANDENBRANDEN (J.), & BEQUAERT (J.). *Essais de Transmission du Trypanosoma gambiense par la Glossina morsitans*. [Attempts to transmit *Trypanosoma gambiense* by *Glossina morsitans*.]—*Bull. Soc. Path. Exot.* 1912. Nov. Vol. 5. No. 9. pp. 762-770.

Brief details are given of four series of experiments carried out with *Glossina* bred in the laboratory.

In the first series the flies were fed upon human patients, in the second upon a goat infected with the human trypanosome, in the third upon a dog and a guinea-pig infected with a strain of trypanosomes from a different source, and in the fourth upon infected monkeys.

From the experiments it is seen that at Sankisia at an altitude of 750 metres the *T. gambiense* can undergo development in *G. morsitans*, and the flies can transmit the infection to animals by biting them.

In two flies which were proved capable of transmitting the trypanosome the salivary glands were found to be invaded with flagellates, but there was no evidence of multiplication in the proboscis. In some cases typical trypanosomes were found in the liquid in the hypopharynx, but the authors think that these had escaped from the salivary glands. They believe that these represent the only infective form.

At Sankisia only 1·7 per cent. of the experimental flies were found to be infected, and the only flies to become infective were those fed upon the infected monkeys. The authors think that this may be explained in part by the fact that the trypanosomes were always numerous in their blood.

The period elapsing between the infective feed and time at which the flies became infective was in one case less than 24 days, and in the other two 30 and 35 respectively.

Although the experiments shew that *G. morsitans* is capable of transmitting *T. gambiense* the authors have been unable to obtain any direct evidence that these flies play any part in the transmission of the trypanosomiasis which is responsible for large numbers of deaths in Katanga. They have found villages where only *morsitans* occurs, but they are not certain whether the inhabitants have not visited areas where *palpalis* is present, such areas being not far distant. The authors have not been able to make experiments with wild flies because a large proportion of the flies are infected with *brucei* and *congolense*, and a larger number of animals than were at their disposal would be necessary.

- (297) BLANCHARD (M.). *Marche de l'Infection à Schizotrypanum cruzi chez le Cobaye et la Souris*. [The Course of Infection with *Schizotrypanum cruzi* in the Guinea-pig and the Mouse.]—*Bull. Soc. Path. Exot.* 1912. Oct. Vol. 5. No. 8. pp. 598-599.

Two infected guinea-pigs were originally received by the author. The blood of these animals was found to contain only a very few parasites at the time of receipt, but there was a gradual increase in the number present until the 30th day, after which the number

diminished until by the 44th day none could be found. One of the guinea-pigs was subjected to four subsequent intraperitoneal inoculations with blood containing a large number of parasites, but failed to become reinfected. Two young born of this animal were found to be as susceptible to infection as control animals.

Trypanosomes were not discoverable in the blood of the second guinea-pig after the 44th day. It was killed on the 67th day and its blood used for the inoculation of two fresh animals. One of these became infected, and the other resisted infection, although it was subsequently proved to be susceptible by a positive inoculation. The guinea-pigs shewed no clinical evidence of infection.

Three other guinea-pigs inoculated in series from the second of the two original animals became infected, and the course of the infection was practically the same in two of the cases. The period of incubation was 9 to 13 days, and the parasites increased in number in the blood up to the 34th and 43rd days. The third guinea-pig shewed parasites in its blood on the 8th day after inoculation and they became numerous by the 16th day. During the next eight days there was a rapid decrease in the number present, but they did not completely disappear until the 85th day. Subsequently they reappeared and increased in number until the time of death on the 127th day.

Seven passages have been made in mice, and the course of the infection has been very constant, and it has always terminated fatally. The period of incubation in this animal is 6 to 7 days. The parasites rapidly increase in number in the blood and remain numerous until death about the 15th to 20th day.

(298) BRUMPT (E.). *Pénétration du Schizotrypanum cruzi à travers la Muqueuse Oculaire Saine.* [Penetration of *Schizotrypanum cruzi* through Healthy Conjunctiva.]—*Bull. Soc. Path. Exot.* 1912. Nov. Vol. 5. No. 9. pp. 723-724.

The most susceptible monkey to infection with *Schizotrypanum cruzi* is *Cercopithecus ruber*. The author placed some faecal matter from an infected *Conorhinus megistus* upon the eye of one of these monkeys and a fatal infection ensued.

The faeces of *Conorhinus* and *Cimex* are very virulent and contain very mobile slender trypanosomes which have a great power of penetrating into tissues.

Experiments in which the faecal matter was placed upon the skin of rats and monkeys had negative results.

SPIROCHAETOSIS.

(299) GASPERI (F. de). *Présence d'un Spirochète dans le Sang d'un Cobaye.* [The Presence of a Spirochaete in the Blood of a Guinea-pig.]—*Bull. Soc. Path. Exot.* 1912. Oct. Vol. 5. No. 8. pp. 589-591. With 1 text-figure.

The spirochaete here described was encountered in cultures made from the heart blood of a guinea-pig which, in the course of other experiments, had been inoculated with garden earth which had been ground in mortar with water at 85° C.

The cultures were made in glucose agar and incubated at 42° C., and the spirochaete was found in the water of condensation on the day following the preparation of the cultures.

The organisms varied from 16 to 24 microns in length, and measured 0.3-0.4 in width. They showed five to eight turns, and the ends were distinctly tapering. The spirochaetes died after about forty-eight hours, and the author was thus prevented from isolating it.

With the object of ascertaining whether this spirochaete was in the nature of an accidental invader the inoculation experiments were repeated with similar materials ten times, but in no case was a spirochaete found.

The author concludes that the spirochaete is a specific organism of the guinea-pig.

- (300) MARCHOUX (E.) & COUVY (L.). *Argas et Spirochètes*.—*Bull. Soc. Path. Exot.* 1912. Dec. Vol. 5. No. 10. pp. 796-798.

Referring briefly to the investigation of various authors regarding the presence of "granules" in the bodies of ticks infected with spirochaetes, Marchoux and Couvy state that they have examined *A. vespertilionis*, *R. ricinus* and *L. echidninus* and have found the "granules" in each species and in each individual, although the ticks had not been fed upon animals affected with spirochaetes. They conclude that the "granules" have no connection with spirochaetes. They propose to show in a future communication that in *Argas* the spirochaetes behave like bacteria and do not undergo any cycle of development comparable to those seen in intermediary hosts among the protozoa.

- (301) MESSERSCHMIDT (T.). *Die chemotherapeutische Beeinflussung der Hühnerspirochätenkrankheit, durch die im Handel befindlichen Jodpräparate*. [The Effects of Commercial Compounds of Iodine in Spirochaetosis of the Fowl.]—*Zeitschr. f. Immunitätsforsch. u. Exp. Therap.* 1. Teil., Orig. 1912. Nov. 2. Vol. 15. No. 2-3. pp. 293-302.

In these experiments a number of different preparations of iodine were used. None of them was found to have any toxic effect nor was any of the slightest use either for preventive or curative treatment.

- (302) PATTON (W. S.). *Spirochaeta ctenocephali*, sp. nov., parasitic in the Alimentary Tract of the Indian Dog Flea, *Ctenocephalus felis*.—*Ann. Trop. Med. & Parasit.* 1912. Oct. 18. Vol. 6. No. 3. B. pp. 357-359.

In view of the recent work of BASILE and others regarding the development of the parasite of canine kala azar in the dog flea—*Ctenocephalus canis*—importance attaches to the study of the parasites natural to the dog flea, and particularly in India where canine kala azar has not as yet been observed. The common dog flea in Madras is *C. felis*, *C. canis* having been found so far on the jackal only.

The parasite described in this paper has been found both in the larva and in the adult flea. Out of a total of some 1500 larvae the parasite was found in two only derived from fleas fed upon cats kept at the laboratory, and it was found in one adult flea obtained from a dog out of 500 fleas examined. The author proposes the name *Spirochaeta ctenocephali*.

- (303) FONTANA (A.). *Metodo per Colorare Intensamente e Rapidamente il Treponema pallidum ed Altri Spirocheti*. [A Quick Method of Staining Spirochaetes intensely.]—*Pathologica*. 1912. Oct. 1. Vol. 4. No. 94. pp. 582-583. With 2 text-figures.

The material for examination is diluted with a drop of distilled water, spread on a slide, allowed to dry in the air, and then fixed by heat. Cover the preparation with 5 per cent. tannic acid in distilled water and warm until vapour arises (20 secs.). Wash in tap water for 30 seconds. Pour on the slide a few drops of ammoniacal solution of silver nitrate (prepared by adding ammonia to a five per cent. solution of silver nitrate in distilled water until the precipitate which is first formed is just redissolved) and warm for 20 to 30 seconds. Wash and dry. The spirochaetes are stained from an intense yellow to a deep brown.

LEISHMANIASIS.

- (304) NICOLLE (C.) & BLAIZOT (L.). *Virulence des Cultures de Leishmania infantum. Sensibilité du Chacal au Virus du Kala Azar Tunisien*. [Virulence of Cultures of *L. infantum*. Susceptibility of the Jackal to the Tunisian Virus.]—*Bull. Soc. Path. Exot.* 1912. Nov. Vol. 5. No. 9. pp. 721-723.

Having lost their human strain of *Leishmania* which had been preserved by passage since 1907, the authors attempted to renew it by inoculating animals with cultures derived from the same strain. Their first attempt, in which twenty-five cultures of the forty-fifth generation were used for the intraperitoneal inoculation of a dog, failed to infect.

A fresh series of experiments was then instituted, a more recently isolated Tunisian strain being employed. A number of rabbits and guinea-pigs were inoculated without result.

A puppy, about a fortnight old, was inoculated intravenously with fifteen cultures of the same generation sixteen days old. The inoculation was followed by immediate prostration with marked dyspnoea; the animal recovered, however, after some hours. When the dog was killed on the 72nd day parasites were found to be fairly numerous in the spleen and very numerous in the bone-marrow. A second dog inoculated from the first was found to be infected when it was killed on the 82nd day. A jackal, also inoculated with material from the first dog, when killed on the 84th day was found to be infected and parasites were found to be fairly numerous in the marrow, scanty in the spleen, and absent from the liver. The inoculations were intraperitoneal, but the exact material used is not stated.

- (305) LAVERAN (A.). Infections des Souris et des Rats dues au Kala-azar Méditerranéen et au Kala-azar Indien. [The Infection of Mice and Rat with Mediterranean and Indian Kala-azar.]—*Bull. Soc. Path. Exot.* 1912. Nov. Vol. 5. No. 9. pp. 715-721.

After a brief reference to previous successes obtained by himself and YAKIMOFF, the author gives details of experiments made with twenty-six mice.

The strain used was that occurring in Tunis, and the materials were obtained from dogs and mice.

The mice were inoculated intraperitoneally, and spleen-pulp, liver, and bone-marrow were used. In four cases there was severe generalised infection, in four instances the infection was generalised but moderate, and in thirteen it was generalised but slight. In the remaining five mice the infection was only localised, or there had been complete recovery before the animals were killed.

Severe infections were found in mice killed twenty-four and thirty-one days after inoculation. In a third mouse this condition was found when it was killed on the 138th day, but the author considers this to be exceptional. In most of the mice killed at about this time the infection was only a slight one, and the animals appeared to be on the way to recovery. One mouse died after having been inoculated three times, twice with culture, and once with the spleen pulp of a heavily infected mouse. The enlarged spleen was found to contain large numbers of the parasite, and there were no lesions to account for the animal's death, but the author does not feel justified in attributing it to kala azar, as he has found more severe infections in mice which survived.

Eight days after inoculation the parasites were found to be more or less numerous present in the peritoneal exudate. Many were found within mononuclear leucocytes and endothelial cells, but free forms also occurred. In some instances the infection remained confined to the peritoneum, but in other cases the liver, spleen, and bone-marrow were invaded.

Contrary to what might have been expected, it was not easier to produce infection with cultures containing flagellates. It was found that the flagellates were rapidly ingested by phagocytes, while the non-motile forms of the organism were able to live and multiply within the leucocytes and the endothelial cells.

Only negative results were obtained when mice were inoculated intraperitoneally with cultures of *L. infantum*. In one instance a slight infection was produced by intravenous inoculation with this material.

A series of passage experiments showed that there was no exaltation of virulence of *L. infantum* for the mouse, but rather the reverse. In infected mice there is a variable amount of enlargement of the spleen, up to six times the normal, and the parasite is most frequently found in that organ. In one instance in which smears from the spleen were negative, the inoculation had been intra-hepatic instead of intraperitoneal as was generally the case.

In rats the course of the infection is much the same as in mice. In one rat which died twelve days after inoculation (method not specifically stated) the parasite was present in fairly large numbers in the peritoneal exudate, but was very scantily present in the enlarged spleen.

In view of the fact that the different behaviour of the Mediterranean and Indian strains in dogs has been put forward as evidence contra-indicating their identity, the author advised Row to carry out experiments with the Indian strain.

Row states that he has succeeded in producing generalised infection in two mice by inoculating them intraperitoneally with material obtained from subcutaneous nodules produced in a *Macacus sinicus* with the human strain of Indian kala azar.

Mention is made of PATTON's success in infecting a rat with material from a person dead of Indian kala azar, and reference is made to PATTON's view that the two are not identical.

It is admitted that there is not sufficient evidence upon which to base any definite view, but it is pointed out that although the Indian strain appears to be the rather more virulent for the mouse, both are capable of producing generalised infection.

- (306) SERGENT (Edm.), SERGENT (Et.), LHERITIER (A.), & LEMAIRE (G.). *Transmission de Leishmania de Chien à Chien par Piqûres de Pulex serraticeps*. [The Transmission of Leishmania from Dog to Dog through the Bites of *Pulex serraticeps*.]—*Bull. Soc. Path. Exot.* 1912. Oct. Vol. 5. No. 8. pp. 595-597.

I. A young bitch in good condition was subjected to the bites of fleas (*P. serraticeps*) on eighty-two occasions, the average period of biting being five minutes. Each flea used had previously been fed upon an infected dog. Before the experiment was commenced the dog was under observation for two months and appeared healthy. Liver punctures were negative. During the course of the experiment the dog was very carefully protected against and kept free from all ectoparasites.

Rather more than a month later the animal began to lose condition and the clinical symptoms of infection made their appearance, but examination of marrow from the femur was negative.

About two months later the animal began to improve in condition, and was then killed.

At the post-mortem, preparations from the liver were negative, but in smears from the spleen and bone-marrow *Leishmania* was found in small numbers, and apparently in a degenerated condition.

Cultures from the spleen on NNN medium yielded numerous *Leptomonas* after eight days, five out of twelve tubes being successful. The strain had, up to the time of writing passed through thirteen subcultures.

Cultures from the bone marrow were negative.

A control dog kept under identical conditions, save for the biting of the fleas, shewed no infection.

II. A young dog was bitten daily for fifteen days by a single flea which had been fed upon an infected dog every two days during the preceding fortnight. The animal was killed at the end of three months, but no evidence of infection could be detected.

III. A young dog was bitten daily for four days by a single flea which had had a single meal on an infected dog two days previously. The result in this case was also negative.

Microscopic examination of the intestinal contents of fleas were made and the following forms were found:—

In the intestinal contents of two fleas that had fed on an infected dog and in two out of nine fleas fed upon a supposedly healthy dog were found large numbers of resting forms about five microns in diameter. In many cases these were collected together into clumps. They possessed a fairly large nucleus and a blepharoplast.

In the faecal matter from the same fleas, and in the contents of the posterior portion of the intestine, slender, binucleated flagellates, measuring ten to twelve microns in length, were found. Transitional forms were also observed.

One cell was observed containing three parasites in the resting stage.

(307) WENYON (C. M.). Experiments on the Behaviour of *Leishmania* and Allied Flagellates in Bugs and Fleas, with some Remarks on Previous Work.—*Jl. London School Trop. Med.* 1912. Dec. Vol. 2. Part 1. pp. 13-26.

This paper contains a criticism of the view held by PATTON that the bed bug is the true host of *Leishmania*, and much evidence is adduced to indicate that this is not the case. The author gives evidence which he has obtained indicating that it is not improbable that *Stegomyia fasciata* is the true host of the parasite.

Experiments carried out by the author on himself with *Cimex lectularius* indicated that no multiplication of the parasite took place in the only three bugs that were found to contain a small number of flagellates, out of a total of a hundred and five that were fed.

In a further series of experiments attempts were made to infect human and dog fleas, but in no instance was any development of the parasite discovered.

During the course of these experiments it was found that fleas infected with *T. lewisi* remain infected for long periods whether they feed upon human or rat blood, and that the trypanosome appears to undergo its development in *Pulex irritans* as well as in the dog flea, *Ctenocephalus canis*.

Nöller's observation that it is the ingestion by the rat of infected faeces from the flea that is responsible for the transmission of the trypanosome was confirmed. This was done by feeding a dog flea on an infected rat, subsequently feeding it upon a second

rat, and giving the faeces passed during the first feed to a third rat. Only the third rat became infected.

The author thinks that BASILE and others may have been mistaken in their view that they had traced multiplication forms of *Leishmania* in the flea, owing to the fact that the flea contains a natural herpetomonas almost indistinguishable from cultures of *Leishmania*, and no steps were taken in his experiments to exclude this possibility. The author believes, with NICOLLE and BASILE, that the Mediterranean disease in dogs and children is the same.

(308) LAVERAN (A.). *Présentation de Macaques inoculés avec succès au moyen d'une Culture de la Leishmania du Bouton de Delhi.* [Monkeys successfully inoculated with Cultures of the *Leishmania* of Delhi Boil.]—*Bull. Soc. Path. Exot.* 1912. Oct. Vol. 5. No. 8. pp. 573-575. With 1 plate.

The original material was derived from some cultures received from Row at Cambay. No motile flagellates were discoverable in the cultures on their arrival in Paris, but subcultures were obtained on the Novy-Nicolle medium. The strain was kept alive in the laboratory by sub-inoculating about every fortnight, the tubes being incubated at 22° C.

The author has obtained the best results by inoculating intradermally. He has not been able to produce infection by inoculation into mucous membranes.

The period of incubation is variable, but the minimum observed was fourteen days. The lesions take the form of little intradermic indurations at the seats of inoculation which vary in size up to that of a pea. There is no inflammatory congestion, nor pain on pressure. When the lesions attain the size of peas they come to a head, and if they are incised they are found to contain a droplet of thick whitish liquid.

Nodules which are not incised become covered with small brown crusts which are easily detached leaving the ulcerating tissues beneath exposed. In some cases the nodules are reabsorbed.

Parasites are scantily present in preparations made from the crusts, or from the sero-sanguinolent liquid beneath them, but they may be numerous in preparations made with material scraped from the depth of the ulcers. The great majority of the parasites appear to be free, but this may be due to the destruction of the host cells during the preparation of the films. The parasites present the characters of *L. tropica*. The nuclei are rounded and not flattened against the side of the parasites as is often the case in *L. americana*.

In one animal the ulcers had not healed after 74 days.

The author states that his results do not differ greatly from those obtained with the Tunisian virus by NICOLLE and MANCEAUX. He thinks that there is no specific difference between the parasites of Delhi Boil and Gafsa Boil (Tunis).

The author has been unable to infect dogs and mice with cultures of the Indian strain of *L. tropica*.

- (309) Row (R.). Some Experimental Facts re Kala-Azar (Indian).
—*Jl. of Trop. Med. & Hyg.* 1912. Nov. 1. Vol. 15.
No. 21. pp. 327-328. With 2 text-figures.

In the present paper some account is given of the results obtained by infecting *Macacus sinicus* with original virus and an old culture derived from the original virus.

The virus was obtained from a human source by splenic puncture and was fairly rich in parasites. A monkey was inoculated by scarification with some of this original material, and nine weeks later small pin-head nodules developed at the two seats of inoculation. In preparations made from material squeezed from these lesions the parasites were recognisable, and after a period of six weeks were still to be found, but at this time they were not quite so typical as those found in the first instance.

A second monkey was inoculated subcutaneously with a fifth subculture from the same case. The culture was full of rounded forms a fortnight after it had been observed to be rich in flagellates. The doses injected were 0.3 and 0.15 cc. Nothing was observed until five months after inoculation when small hard nodules were found under the skin at the seats of inoculation. Preparations were made from material obtained by puncture and a fair number of parasites like those found in the spleen were found. In culture these parasites were found to be present in the preflagellate stage at 24 hours, and flagellates were observed on the fourth day.

VARIOUS BLOOD PARASITES.

- (310) CARINI (A.) & RUDOLPH (M.). Sur quelques Hématozoaires de Lézards au Brésil. [Some Haematozoa of Lizards in Brazil.]
—*Bull. Soc. Path. Exot.* 1912. Oct. Vol. 5. No. 8.
pp. 592-595. With 10 text-figures.

1. *Haemogregarina ameivae*.—This parasite has been encountered in a number of specimens of *Ameiva surinamensis*. It occurs in the peripheral blood, in the interior of the red cells.

In the living state it appears as an elongated clear space with an irregular outline. No movement is exhibited.

In preparations stained with Giemsa the protoplasm is found to be very delicate, scarcely stained at all, and always rather shrivelled. The parasite is elongated in shape and somewhat irregular in outline. The ends are rounded, and one may be larger than the other. The nucleus is oval in shape and is situated towards the smaller end of the parasite. It occupies practically the whole width of the body and stains of a violet tint.

The organism measures 11 to 13 microns in length by 3 to 4 in width. It possesses a capsule which is not distinctly visible in the endoglobular forms, but is quite distinct in the extra-globular forms encountered in smear preparations made from the internal organs.

As a rule the parasite lies in the centre of the red cell causing displacement of the nucleus, which is often associated with some enlargement and deformation.

In spite of careful examinations of preparations from a number of lizards no multiplication forms have been seen. In a single instance a number of clear regularly oval spaces were seen in the liver which had the appearance of being empty cysts.

2. *Plasmodium minasense*.—This organism is an endoglobular pigmented parasite, and the authors have encountered it in small numbers in stained preparations on one occasion only.

The parasite which is rounded or oval in shape is situated at one of the poles of the red corpuscle invaded by it, the cell showing little or no alteration in shape. Parasites of various sizes were seen, the largest measuring 4-5 microns.

The protoplasm stains blue, and contains a number of small vacuoles. Granules of pigment are scattered through the cytoplasm. The granules are irregular in shape and size, and of a brownish-black colour. In some cases they are scattered through the protoplasm in a disorderly fashion, and in others they are collected together into a single mass.

The nucleus is composed of granules of chromatin which stain of a pale pink colour.

Certain details of structure suggested that the parasites seen were male and female gametes, but the number found was so small that no definite opinion as to this could be formed.

A small number of dividing forms have been seen. In these cases the number of merozoites was very small.

3. *Trypanosoma species?*—This organism has also been studied in stained preparations only. The parasite is stumpy, but somewhat variable in shape. In every specimen the protoplasm was traversed by clear streaks which were not constant in position. The cytoplasm stained a fairly intense blue colour and appeared to be finely granular. The nucleus was oval in shape and measured 5 by 3 microns. It occupied a position about the middle of the body and stained uniformly pink.

The blepharoplast was situated close to the nucleus, was oval in shape, and was surrounded by an unstained zone. In more intensely stained specimens a delicate filament could be seen starting at the blepharoplast, passing along a very poorly developed undulating membrane, and terminating in a short free flagellum.

The parasite appears to be related to the *T. perroteti*, and the name *T. rudolphi* is suggested for it.

- (311) LEGER (A.). Présence de deux Leucocytozoaires morphologiquement distincts dans le Sang du Chien, à Bamako (Haut-Sénégal et Niger). [The Presence of two Morphologically Distinct Leucocytozoa in the Blood of the Dog at Bamako (Upper Senegal).]—*Compt. Rend. Soc. Biol.* 1912. Oct. 25. Vol. 73. No. 29. p. 376.

The author mentions the occurrence in the dog of the leucocytozoon previously recorded as occurring in the spotted hyena (*Hyena crocuta*) (see Ref. 100, this *Bulletin*. Vol. I. No. 1. p. 58). The *Leucocytozoon canis* has also been found, but double infection has never been observed in the same animal. The new

leucocytozoon is described as being smaller, and more stumpy than *L. canis*. Two dogs were found to be infected out of 114 examined. The parasite invades mononuclear leucocytes only, and no free forms have been observed. The name *Haemogregarina chattoni* is suggested.

(312) YAKIMOFF (W. L.) & KOHL-YAKIMOFF (Nina). *Toxoplasma canis* (Mello).—*Archiv f. Protistenkunde*. 1912. Vol. 27. pp. 195-206. With 2 plates.

The first section of this paper is devoted to an account of the discovery by various authors of *Toxoplasma* in a number of animals. NICOLLE and MANCEAUX discovered the parasite in a rodent (*Ctenodactylus gondi*) in Tunis, SPLENDORE in the rabbit in Brazil, MELLO in the dog in Italy, PROWAZEK in a mole from Japan, and CARINI in the pigeon in Brazil.

According to NICOLLE and MANCEAUX, *Toxoplasma* is curved in outline, with pointed ends, but occasionally round forms may be found. As a general rule the parasite discovered by these authors measured about five microns in length by three in width, but larger forms up to seven microns in length were observed. The parasite showed no motility. The cytoplasm was alveolar in structure. The nucleus was rounded or oval and measured about two to three microns. Multiplication is said to take place by longitudinal division. Both free and intracellular forms were seen, the latter occurring most frequently in lymphocytes. The majority of the parasites were found in the spleen, liver, and mesenteric glands; they were present in smaller numbers in the lungs and kidneys, and exceptionally in the heart blood and bone marrow.

These authors succeeded in transmitting the parasite to one out of twelve guinea-pigs inoculated, but they were unable to obtain cultures on either Novy's or Nicolle's blood-agar. They were unable to infect apes or rats.

According to SPLENDORE, affected rabbits show no symptoms save wasting and, in some cases, some paralysis of the hind legs just before death. At the post-mortem the liver and spleen were found to be enlarged and besprinkled with greyish-white points. In many cases there was ulceration of the intestinal mucous membrane, and a large quantity of serous fluid in the body cavities.

SPLENDORE's organism was rather longer and narrower than that described by NICOLLE and MANCEAUX, and in addition to the curved forms this author found cyst phases containing either free parasites or parasites in a condition of irregular division. He also encountered round or amoeboid forms. There was a variable amount of compact cytoplasm which stained of an intense blue colour with Giemsa.

SPLENDORE believes that multiplication takes place both by longitudinal division and by schizogony. He succeeded in transmitting the parasite to other rabbits, guinea-pigs and birds; he failed however to infect dogs, and was unable to cultivate the parasite.

According to SPLENDORE, CARINI succeeded in infecting rabbits, guinea-pigs, white rats, and pigeons with *Toxoplasma*, the latter giving the best results. He failed to transmit the infection to the dog.

CARINI agrees with SPLENDORE regarding the methods of multiplication.

Toxoplasma in the dog was first discovered in Italy by MELLO. The parasite was found in all the organs, but principally in the liver and lungs. *Toxoplasma* was exceedingly numerous in the ulcerated intestine.

In smear preparations from the liver the parasite resembled *T. gondi* described by NICOLLE and MANCEAUX, save that it was very much smaller, measuring only 2 microns. Free and intracellular forms were seen, and in many instances as many as 60 individuals were found in a single cell.

CARINI was able to infect pigeons from dogs infected with *Toxoplasma*. In one case the infection was successfully transmitted to another dog, but a second dog failed to become infected, as did also rats, sheep, horses and cows.

CARINI is of the opinion that the *Toxoplasma* occurring in the dog in Brazil is not identical with that found by MELLO, on account of the great discrepancy in size. He considers it to be identical with that found in the rabbit.

The present authors encountered *T. canis* in a dog that was under experiment with a strain of *Leishmania infantum* derived from Tunis. The greater number of parasites were found in the bone-marrow and spleen, infection being less extensive in the liver, and still less in the lungs. Parasites were found once only in the blood.

The parasite occurs typically in a crescentic form, one end being pointed, and the other more or less rounded. The cytoplasm stains of a blue colour to a varying depth with Giemsa, and is alveolar in structure. Many of the organisms contain vacuoles. The nucleus stains red or reddish-violet, is oval or more rarely rosette shaped, and contains either a darkly staining caryosome or several chromatin granules. In many specimens the cytoplasm at one end stains reddish, and in a few of the parasites red granules are present at one or other end. The appearances closely resemble those shown by some haemogregarines and sarcosporidia.

In addition to the crescentic forms there are oval forms, the cytoplasm of which is honey-combed in appearance. The nucleus is irregular in outline and often fragmented, and is sometimes set at right angles to the length of the parasite.

In dividing forms, of which a large number were seen, the nucleus appears, at least in dry-fixed specimens, to divide amitotically, division of the cytoplasm taking place longitudinally. Many intermediate forms between the crescentic and oval forms were seen.

In the schizonts and encysted forms a large number of nuclei were observed. The number varied from two to sixty, and the fragments developed into crescentic and oval forms. During the process of schizogony residual bodies which stained deep blue were formed.

The intracellular parasites were enclosed in mononuclear lymphocytes, or neutrophile polynuclears, and never more than two schizonts were seen in a single cell. In two instances the parasite was found enclosed in liver cells.

The authors are of the opinion that the *T. canis* is a parasite specific for the dog.

With regard to the classification of the parasite, the authors agree with PROWAZEK that it should be classed with the Haemogregarina among the Leucocytogregarina.

Infection experiments failed as the infected dog had been dead two days when the experimental inoculations were made, and the parasites were then dead.

The authors do not think that the infection was brought from Tunis in the dog infected with *L. infantum* for the following reasons:—Toxoplasma was not found in smears from any of the organs of the imported dog. Of the eight dogs inoculated from the African dog this one only showed the Toxoplasma infection, and they cannot suppose that this dog was alone susceptible. The dog infected with Toxoplasma was from two to three months old and was born in Germany. They therefore think that it was a case of spontaneous infection.

- (313) CARINI (A.). Sur un Nouvel Hématozoaire du Pigeon. [A New Haematozoon of the Pigeon.]—*Compt. Rend. Soc. Biol.* 1912. Oct. 25. Vol. 73. No. 29. pp. 396-398. With 5 text-figures.

The parasite described in this paper was only encountered once, and owing to an accident a bird inoculated with it died without having become infected.

At first sight the author thought that the organism was Haemoproteus, and in consequence of that the examination of living preparations was not very thorough.

The parasite is practically always found in the interior of red corpuscles, of which it causes neither enlargement nor decolourisation. When the organism is large the nucleus is pushed to one side. Some of the corpuscles which contained no parasites were observed to have a speckled appearance. Active movement of the parasite within the corpuscles was not observed.

In preparations stained by Giemsa or Leishman the protoplasm stained of a blue or violet colour, and often contained vacuoles and brown granules. The pigment was disposed towards the periphery or the poles of the elongated organisms.

The nucleus was represented by a small homogeneous mass with indistinct contour, and stained pink.

Not uncommonly two or more parasites were found in the same corpuscle.

The following forms were observed:—

Small forms measuring only 2.3 microns. These forms were rounded, and their protoplasm stained more intensely at the periphery than at the centre. Traces of pigment could be observed in these.

Rounded or oval intermediate forms resembling the small ones except in size.

Dumb bell forms closely resembling the gametes of *Halteridium*. The largest of these measured from 9 to 11 microns in length, by 4 to 5 in width.

Forms provided with pseudopodia. Pseudopodia were observed in rounded, oval, and dumb bell forms. The pseudopodia varied from mere projections to long slender processes exceeding the parasite in length.

Careful examination of preparations from the spleen, liver, lungs, kidneys, and bone marrow failed to reveal any multiplying forms.

The name *Plasmodium columbae* is suggested for the parasite.

- (314) LEGER (M.). *Présence de Haemogregarina canis en Corse.* [The Occurrence of *Haemogregarina canis* in Corsica.]—*Compt. Rend. Soc. Biol.* 1912. Dec. 13. Vol. 73. No. 35. pp. 617-618.

After a brief reference to the discovery of this parasite by different authors in various tropical countries, and to BASILE'S discovery of it in a dog in Rome, the author describes its occurrence in one dog out of twenty examined in Corsica. The animal was also the host of *F. immitis*, but the infestation was a slight one.

In preparations of blood stained by Giemsa the leucocytozoon was found to be invariably intracellular, and enclosed in a capsule which to some extent resisted the penetration of the stain.

The cyst was oval and measured from 10 to 12.5 microns in length, by 4 to 6 in width. The ends were similar. The nucleus was relatively large, and took the form of a slightly curved rod staining of a bright pink colour.

The author has encountered forms resembling those described by WENYON, the parasite being folded upon itself in the form of a U with unequal arms. The protoplasm of the parasite was granular.

The host-cells were mononuclear leucocytes. In some cases the nucleus was fragmented, the fragments tending to stain less intensely.

HELMINTHS.

- (315) MASON (E.). *Larvae of Linguatula taenioides in Camels.*—*Dept. of Public Health (Veterinary Section), Cairo. Paper No. 5.* 1912. p. 10.

"A very high percentage of camels are found to harbour in their mesenteric lymphatic glands the larvae of *Linguatula taenioides*. The glands are found to be hyperaemic and enlarged, or are oedematous, and contain cavities. The parasites are particularly numerous in the cavities, and in these cases are also found free in the peritoneum. The discovery of this parasite (which has been signalled in India) in camels probably explains to some extent the frequency with which peritonitis occurs in this species. The parasites do not themselves produce the peritonitis, but by destroying the mesenteric glands favour the entrance of bacteria."

- (316) MAUPAS (E.) & SEURAT (L. G.). Sur un Nématode de l'Intestin Grêle du Dromadaire. [A Nematode of the Small Intestine of the Dromedary.]—*Compt. Rend. Soc. Biol.* 1912. Dec. 20. Vol. 73. No. 36. pp. 628-632. With 10 text-figures.

Nematodirus mauritanicus. n. sp. The worm is small and slender. It is provided with a thick cuticle which at the anterior end is doubled, forming a slight vesicular swelling, which is transversely striated. In the post-vulvar region the cuticle is formed of two layers, the inner of which is thick and homogeneous, and the outer very thin and striated transversely. There is no evidence of longitudinal striation.

Anterior to the vulva the cuticle is quite soft and devoid of all striation for a distance of 2 to 3 mm. Anterior to this longitudinal striation appears and is continued forwards to the point where the cuticle is double, transverse striation appearing at this point.

The mouth opens in a short funnel-like depression and leads to a straight oesophagus which shews considerable swelling in its posterior part.

The nerve-ring is situated just behind the middle of the oesophagus. The excretory pore opens behind the junction of the oesophagus with the intestine. The cervical papillae are very small.

The female measures from 21 to 24 mm. in length. Posteriorly the female is truncated, but carries a short delicate prolongation. The anus is situated 105 microns from the truncated end, and the vulva a little posterior to the middle point of the body.

The short straight vagina leads to two sacculs of unequal size.

The eggs contained in the uterus are arranged in two parallel rows in the posterior portion of the body. The eggs are large, and measure 220-280 microns, and eggs containing larvae are found side by side with eggs in the morula stage.

The male varies from 13 to 15 mm. in length.

The caudal bursa is composed of two large lateral wings and a small posterior wing which is divided into two portions by a wide gap, each part being supported by a ray which is bifid at its extremity.

The two spicules are brown in colour and very slender. They measure about one-third of the length of the parasite and are fused together by means of a thin membrane for six-sevenths of their length.

- (317) BAUCHE (J.) & BERNARD (P. N.). Note sur quelques Filarioses Animales de l'Annam Central. [Animal Filariases in Central Annam.]—*Bull. Soc. Path. Exot.* 1912. Oct. Vol. 5. No. 8. pp. 622-624.

Subcutaneous filariasis of the dog.—The occurrence of this parasite in two cases, identified by RAILLIET and HENRY, has already been recorded by the authors, but they have now encountered thirty infected dogs. The parasite—*Dirofilaria repens*—was found in thirty out of a hundred dogs taken at random. In each

animal from 8 to 30 specimens were found, the females outnumbering the males in the proportion of two to one.

The authors state that examination of the immature parasites, of the adults, and of their natural habitat enables them to assert that the parasite is not the *Dirofilaria immitis*. There can be no doubt that subcutaneous filariasis of the dog is a pathological entity, special symptoms characterise the affection, and a diagnosis can be based upon the characters presented by the embryos in the circulating blood. All attempts at treatment have failed.

Microfilaria of the pig.—This parasite was encountered in the blood of a young pig during the authors' observations on the peritoneal and pulmonary filariae of the pig. Although hundreds of animals have been examined they have not encountered it a second time.

In the fresh state the parasite measures about 100 microns in length by 4 in breadth. It is very active, but the movement does not involve much translation. The parasite does not possess a sheath. The cephalic extremity is bluntly rounded. The body tapers slightly in its posterior fifth towards the caudal extremity which is also rounded.

In preparations stained with haematin and eosin the average length is about 60 microns, and the body shews three constant specialised parts: 1, a cephalic portion in the form of a V with the apex directed backwards; 2, a ring around the worm practically at right angles to the long axis of the body at a distance of 26 microns from the anterior end; 3, an oval area situated 37 microns from the anterior extremity. The authors have not observed the adult form of the parasite.

Setaria labiato-papillosa (Alessandrini, 1838). This parasite is practically constantly present in the peritoneal cavity of cattle and buffaloes slaughtered for food at Hué, each animal harbouring from 2 to 20 specimens. The young form of the parasite is often found in the anterior chamber of the eye of the horse.

Setaria equina (Abildg. 1789). This parasite is practically always present in the peritoneal cavity of horses.

Ocular filaria of the fowl. *Oxyuris mansonii* is very commonly found in the conjunctival cul-de-sac of the fowl, but it does not appear to cause any symptoms.

- (318) RAILLIET (A.), HENRY (A.), & JOYEUX (C.). *Sur deux Trématodes de Primates*. [Two Trematodes of the Primates.] —*Bull. Soc. Path. Exot.* 1912. Dec. Vol. 5. No. 10. pp. 833-837. With 1 text-figure.

After giving a list of the trematodes, 6 species, already discovered in the Primates, with a brief account of the characters of each, the authors pass to the description of (1) a new species and (2) a parasite which has been described as occurring in man only.

Eurytrema brumpti n. sp. About three hundred specimens of this parasite were collected from the hepatic and pancreatic ducts of an adult female chimpanzee from the Congo.

The body of the parasite is leaf-shaped, greyish-white in colour, and speckled with black and brown. It measures from 3.5 to

4 mm. in length by 1.8 to 2.3 in breadth. The oral sucker is subterminal and opens on the under surface. The ventral sucker is placed at the junction of the anterior and middle thirds. The cuticle is covered with minute scales. The intestinal branches are simple, narrow, and slightly undulating. They extend through about three-fifths of the length of the body. The excretory apparatus has not been thoroughly examined, but the terminal vessel opens in the middle line at the posterior extremity. The genital pore opens at a point anterior to the bifurcation of the intestine. The testicles are at the level of the ventral sucker and, although widely separated from each other, they lie between the branches of the intestine. The ovary is small, rounded, and placed almost in the middle plane of the body, but is slightly displaced towards one side. The vitelline glands are placed laterally, external to the branches of the intestine, and posterior to the ovary.

The posterior third of the body is occupied by the convolutions of the uterus. The eggs contained within the uterus are provided with a thick brownish shell with an operculum at one pole.

Watsonius watsoni (Conyngham, 1904). Six specimens of this parasite were found in the intestinal canal of an old female *Cercopithecus callitrichus* from French Guinea.

The body in preserved specimens is greyish-white in colour, oval in shape and thick. The anterior extremity is rather more attenuated than the posterior. The ventral surface is flat, while the dorsal surface is markedly convex. A fixed specimen measured 5.8 mm. in length by 4.2 mm. in width. On the ventral surface about a quarter the length of the parasite from the anterior end there is a prominent papilla, at the summit of which the genital pore opens. The posterior end of the parasite is occupied by the ventral sucker which measures about 2.5 mm. in diameter.

The digestive canal starts at an infundibulum which is not, properly speaking, a sucker. The pharynx has two obvious lateral diverticula. Bifurcation of the intestine takes place opposite the genital pore, and the thick branches reach almost to the posterior sucker.

The testicles, which are small, are placed one in front of the other, about the middle in the body. They are very clearly lobulated and almost ramified. The female generative organs are placed in the middle line about midway between the posterior testicle and the posterior sucker. Eggs were not observed in the uterus, and no doubt the specimens were immature.

(319) MITTER (S. N.). **A Resumé of our Knowledge on the Occurrence of *Gnathostomum spinigerum* in India and its Relation to a Similar Parasite found in Man.**—*Veterinary Jl.* 1912. Dec. Vol. 68. No. 450. pp. 687-690.

The parasite, which was identified by NEUMANN, was found in a tumour in the stomach of a cat. The worm has been recorded as occurring in *Felis catus*, *F. concolor*, and *F. tigris*. According to COBBOLD it has also been found in a tumour in the stomach of a pariah dog. The present author has found it in a leopard.

The male parasite measures about 5 mm. in length, by 0.5 mm. in breadth, the female being about twice that size.

The parasite in the fresh state is reddish in colour. Four portions are recognisable: a well-marked bulbous head or cephalic portion, a constricted neck, a body, and a tail. The body after leaving the neck becomes dilated and forms a ventral bend and is constant in diameter. The tail is rather narrower and blunt-pointed.

The cuticle of the bulb is covered with eight rows of chitinous leaves, which have their posterior edges notched into spines. The anterior portion of the body is thickly covered with similar trident cuticular laminae. In the middle region the leaves are simple and conical. The cuticle in the posterior portion has no appendages. The oral aperture is simple and guarded by two fleshy lips. The oesophagus is highly developed and expanded at its lower extremity.

The tail of the male is spiral and there are eight genital papillae, while that of the female is straight and trilobed.

From a perusal of the literature referring to them the author believes that *G. spinigerum* and *G. siamense* are practically identical.

MISCELLANEOUS.

- (320) BOUET (G.) & ROUBAUD (E.). *Myiase Prévaginale chez la Vache à Chrysomyia (Pycnosoma) megacephala* Fabr., en Afrique Occidentale. Spécificité Parasitaire des Larves Cuticoles de cette Mouche. [Myiasis in the Cow in West Africa caused by *Chrysomyia megacephala*.]—*Bull. Soc. Path. Exot.* 1912. Nov. Vol. 5. No. 9. pp. 737-739.

After passing references to observations by ROVERE and BRODEN of this condition in the Belgian Congo recorded in 1910, the authors state that one of them observed a case at Seguela, Ivory Coast, in 1907. In this case there was a tumour as large as a fist in the right lip of the vulva which contained a large number of larvae. The larvae when fully developed were identified as being those of *Chrysomyia megacephala*, as were those observed in ROVERE's cases. There was a discharge of sanious pus from the orifice, and when the lips were everted there was found an opening as large as a franc-piece leading to a cavity which contained a hundred larvae. The tumour was said to have developed in a fortnight.

Removal of the larvae with the curette and careful washing-out of the cavity with cresyl for a few days lead to healing.

The majority of the larvae were in a comparatively advanced stage of development. When placed in tubes containing dry or moist earth or sand they rapidly developed into pupae. Those in the tubes containing dry earth remained on the surface during the process, but those in the tubes containing moist earth buried themselves to a depth of two or three centimetres.

From these observations it would appear that this fly is a parasite that is specific for cattle. JOYEUX records the occurrence of

the larvae in tumours on cattle in French Guinea. The invasion by the parasite does not appear to be of an accidental nature, and ROVERE's observations on the biology of the parasite appear to support this view.

According to this author the female fly deposits eggs to the number of a hundred on the hair of cattle, attaching them by means of a special mucus-like material. When the eggs hatch out the larvae penetrate directly into the skin and the subcutaneous tissues, producing necrosis and ulceration. The eggs are never laid on pre-existing wounds. The fact that in the present instance the larvae were all in the same stage of development indicates that the eggs were all laid at the same time by a single insect, whereas if the eggs had been laid on a pre-existing wound this would have been less easy to explain.

From the records of the occurrence of this fly it would appear that it is widely distributed in Africa, but like ROVERE, the authors have been unable to capture the fly in the wild state.

(321) HARTLEY (P.). On the Immune Bodies Occurring in Rinderpest Immune-Serum.—*Indian Civil Veterinary Dept. Memoirs*. No. 3. [Period covered 1910-1911.] pp. 216-230.

The author passes in rapid review the work that has been done in connection with the precipitation of various antitoxins by means of chemical substances, and states that his experiments were begun with the object of discovering with which fraction of the proteins in the immune serum the immune bodies are associated.

A number of experiments were carried out with four samples of serum. The serum was dialysed against tap water and distilled water for three days each, the precipitate obtained being shaken up with sufficient salt solution to make up the original bulk of the serum used. The precipitate did not completely dissolve, but formed an emulsion. This emulsion was found to have the same protective effects as the serum in the same doses. The filtrate had no such effect.

Further experiments shewed that if the precipitate were collected and dried *in vacuo* over sulphuric acid, the powder obtained effectively protected against the simultaneous injection of 0.5 cc. of virulent blood. The powder was simply emulsified in salt solution.

The author's conclusions are as follows:—

1. When rinderpest immune serum is dialysed, the immune bodies are precipitated. The filtrate from the dialysed serum fails to protect susceptible animals against the disease.

2. By this process the rinderpest prophylactic can be prepared in the dry condition.

3. The tests of the dry powder which have been carried out show that the loss of immune bodies is inappreciable and that the dried powder, when dissolved or emulsified, and injected into animals, protects against a simultaneous inoculation of virulent rinderpest blood.

4. By this method about half of the total proteids, which take no part in the immunising process, are eliminated.

5. The rinderpest immune bodies differ from diphtheria and tetanus anti-toxins. The former appear to be similar to the insoluble globulins (Euglobulins); the latter are similar to the soluble globulins (pseudoglobulins).

(322) CROSS (H. E.). The Preparation of Anti-Rinderpest Serum by the Injection of Virulent Artificial Peritoneal Fluid.—*Indian Civil Veterinary Dept. Memoirs. No. 3.* [Period covered 1910-1911.] pp. 206-215.

The preparation followed the plan devised by RUEDIGER which was based upon the discovery of ADIL BEY and NICOLLE that the peritoneal fluid is very virulent.

A solution of 0.5 per cent. of potassium citrate was injected into the peritoneum at the height of a reaction resulting from inoculation with virulent blood. It was found that the injection of this solution in the ratio of 1000 cc. per 100 lbs. body-weight caused death in thirty-six hours to eleven days, and it was determined by experiment that the maximum safe dose that could be administered was 800 cc. per 100 lbs. In the preparation of the serum the quantity was fixed at 700 cc.

By strictly comparative experiments it was found that the serum obtained when this artificial peritoneal fluid was used for hyper-immunising was of lower potency than that obtained when virulent blood was used. This was the reverse of what Ruediger found. The artificial peritoneal fluid is not simply a dilution of the normal fluid as was shown by experiment.

Conclusions.

1. By using artificial peritoneal fluid the amount of virulent material for hyperimmunising purposes is considerably increased, and a marked saving in the cost of serum preparation is effected.

2. The serum from animals injected with artificial peritoneal fluid is usually weaker than the serum prepared by the injection of blood.

3. Artificial peritoneal fluid injections are well absorbed and do not produce sloughing.

(323) HOLMES (J. D. E.). Further Testings of the Haemorrhagic Septicaemia Anti-serum and Vaccine.—*Indian Civil Veterinary Dept. Memoirs. No. 3.* [Period covered 1910-1911.] pp. 242-247.

This short paper forms a continuation of a paper published in an earlier number of the same Journal (No. 1). A brief general account is given of the results obtained in the field, and the details of a small number of experiments are added.

The following are the conclusions arrived at regarding the serum:—

1. The serum confers an immediate protection against the inoculated virus.

2. The immunity following an injection of serum was tested and found satisfactory up to four weeks.

3. The injection of a suitable dose of serum protects in not less than 90 per cent. of cases. A small percentage of animals cannot be protected even by large doses of serum.

4. A potent serum gives immunity in doses which vary according to the size and species of the animal—

For cattle the dose varies from 5 cc. to 20 cc.

For buffaloes " " " 20 cc. and upwards.

For ponies " " " 5 cc. to 20 cc.

For mules " " " 5 cc. to 20 cc.

As the serum is innocuous, and as the period of immunity is somewhat lengthened by increased doses, it is recommended that except in cases of young animals and cattle under 300 lbs., the full dose of 20 cc. should be used.

Conclusions regarding the vaccine:—

1. The vaccine is innocuous. In some cases the injection caused a slight swelling at the seat of inoculation which disappeared in a few days.
2. It confers protection against the inoculated virus for a period of four to six weeks.
3. The protection is not immediate. It sets in about four days following the injection of the vaccine.
4. The vaccine protects in about 75 per cent of cases.
5. The dose of vaccine varies from 5 cc. to 10 cc. according to the size of the animal. A like dose appears to be required for cattle, buffaloes, ponies and mules.

(324) BEVAN (Ll. E. W.) & MILLINGTON (T. G.). *Quarter-Evil in Southern Rhodesia.*—*Jl. Comp. Path. & Therapeut.* 1912. Dec. Vol. 25. No. 4. pp. 286-291.

The authors' experiments are divided into three series, and the objects with which they were carried out and the conclusions arrived at in each series are as follows:—

Series 1.—To determine whether the black-quarter vaccine issued by the Bacteriological Laboratory, Pretoria, affords immunity against the form of quarter-evil occurring in Matabeleland.

Conclusions:

- (1) That the Transvaal black-quarter vaccine, in suitable doses, can be safely applied to sheep.
- (2) That this vaccine has the power to modify the reaction due to inoculation with the local virus.

Series 2.—To test whether an animal immunised with Rhodesian vaccine would resist inoculation with a fatal dose of virulent muscle obtained from the Transvaal.

Conclusions:

- (1) A vaccine can be produced from the muscle of an animal dead of the Rhodesian form of quarter-evil.
- (2) Such a vaccine, in suitable doses, can be applied with safety to sheep.
- (3) Such a vaccine will confer immunity against a fatal dose of infective muscle of an animal dead of Transvaal black-quarter.

Series 3.—To determine whether the so-called "struck" or "strike" of Romney Marsh sheep is identical with the quarter-evil of South Africa.

Conclusion:

From cross-immunity experiments it would appear that a close relationship exists between the quarter-evil of various parts of South Africa and the so-called "struck" of Romney Marsh sheep.

It should be noted that these conclusions are based upon the results obtained in a very limited number of experiments.

(325) BLACKLOCK (B.). *The Resistance of Ornithodoros moubata to Various Sheep-Dips.*—*Ann. Trop. Med. & Parasit.* 1912. Dec. 30. Vol. 6. No. 4. pp. 429-433.

In these experiments a number of dips were used and in each case the solution was prepared in the first instance in the manner laid down in the instructions.

A. Animal experiments.

1. Prophylactic.—The skin of animals (generally goats) having been shaved was soaked with the solution to be tested. Ticks were placed upon the animals while the skin was wet and also after it had dried.

Many experiments were made with Cooper's, Savar's, Hayward's Yellow Paste, and MacDougall's dip, to ascertain whether they

prevented ticks from feeding on the animals. In many cases the ticks refused to feed, but as the same was observed with control animals probably the reason lay with the ticks and not with the dip used.

2. Curative.—The ticks were allowed to feed upon the shaved skin of an animal, and, while still feeding, the various dips were applied to them at different intervals after the commencement of the meal, the dips being poured into the feeding glasses and completely covering the parasites. Scarcity of material prevented these experiments from being carried out as completely as was desirable, but it was gathered that the sooner the dip was applied after the commencement of a feed, the more likelihood was there of the ticks becoming loosened, and apparently also of their subsequently dying.

B. In vitro.

In these experiments the ticks were placed in test-tubes and the dips poured on to them, the ticks being prevented from rising to the surface of the liquid by means of a piece of blotting paper. Examples are given showing that a bug was placed in a 1 in 50 solution of Little's dip for four, eight, sixteen, and thirty-two minutes, the parasite being taken out, dried and warmed between each immersion. At the end of the series of immersions it was alive and active. After a period of rest for ten hours it was again immersed for sixty-four minutes; it was still alive and active, but it was found to be dead after an immersion lasting a hundred and twenty minutes.

Using Cooper's dip in the same strength (1 in 50) a tick was found to survive immersion for two hundred and sixty minutes.

The author states that it is well to draw attention to the fact that ticks belonging to *Argasidae* are generally supposed to have greater powers of resistance than other ticks.

The prophylactic and curative properties of Cooper's dip were tried with solutions of the strength recommended and of greater strengths.

The ordinary strength of solution applied to shaved skin did not completely prevent the ticks from feeding, either while the skin was moist, or after drying. When applied to ticks actually feeding it frequently failed to make them release their hold, and in one case a solution of twice the ordinary strength was applied to ticks feeding on a dog fifteen minutes after the commencement of the feed and kept in operation for two and a half minutes. The ticks went on feeding and were alive and active twenty-four hours after removal. In vitro two hours immersion failed to kill ticks, and in one experiment five hours was effective.

Similar results were obtained in Little's dip. In one experiment a 1 in 50 solution was applied to two ticks feeding on a goat, the application being maintained for twenty minutes. At the end of the time both the ticks were fast, and were alive and active after twenty-four hours.

In vitro twice the strength of solution recommended failed to kill ticks after five hours' immersion.

With MacDougall's dip the results were rather variable. Some parasites fed on the treated skin. But in some cases parasites

feeding, especially when they had only just commenced, were dislodged, and some of these died within twenty-four hours.

In vitro ticks survived thirty-two and sixty-four minutes. In one experiment a tick died after two hours' immersion, but others survived five hours.

Savar's dip.—This dip was not effectual as a prophylactic. In curative experiments ticks were killed in thirty minutes.

In vitro ticks survived two hours' immersion, but not five.

Hayward's Paste.—The prophylactic action of this material was not reliable, and, in the case of feeding ticks the application was not effective in every case, parasites surviving a 1 in 100 solution for thirty minutes.

- (326) JASTREMSKY (D.). Zur Frage über die Negrischen Körperchen. [Regarding Negri Bodies.]—*Centralbl. f. Bakt.* 1. Abt., Orig. 1912. Nov. 9. Vol. 67. No. 1-2. pp. 65-68. With 1 plate.

The author's observations were undertaken with the idea of confirming or otherwise the statements of a number of observers who claim to have discovered Negri bodies in the nervous system of normal animals and of animals suffering from diseases other than rabies. He examined Ammon's horn from the brains of 19 perfectly healthy cats, and in six instances found in the protoplasm of the nerve cells bodies somewhat resembling small Negri bodies. In some cases they were scanty and in other numerous. The largest measured 1.4 microns. He does not think that they have anything to do with Negri bodies, principally because these bodies showed a strong affinity for the methyl blue in Mann's stain.

- (327) CARINI (A.) & MACIEL (J.). La Pseudo-rage ou Paralysie Bulbaire Infectieuse au Brésil. [False Rabies or Contagious Bulbar Paralysis in Brazil.]—*Bull. Soc. Path. Exot.* 1912. Oct. Vol. 5. No. 8. pp. 576-578.

As pointed out by the authors, the disease here described appears to correspond exactly with that described by AUJESZKY in Hungary in 1902, and if the two are identical this is the first record of its occurrence outside that country. The disease affects cattle. The symptoms are very characteristic, and the disease is very rapidly fatal, death occurring as a rule in 24-48 hours. The most striking symptom is intense itching of various parts of the body. The irritation is so severe that the animals do themselves serious bodily harm in their efforts to rub and bite the affected parts. In Brazil the disease is known as "Peste de Coçar" indicating the most marked symptom.

The authors have proved the virulence of the nervous system and the blood by intracranial and subcutaneous inoculation respectively, and they have infected rabbits and cattle with material derived from previous experimental cases. In every case the characteristic symptoms were presented. Rabbits died in three days, and cattle in eleven days.

At the post-mortem examinations no lesions of any magnitude were discovered, the only alterations being a few haemorrhagic

points in the nervous system and slight congestion of the mucous membrane of the alimentary tract.

The causal agent has not been discovered, and all attempts to cultivate it have failed.

The disease does not appear to be very rare as reports of its occurrence have been received from a number of different places. In some instances outbreaks are limited to a few animals, but in others the losses have been considerable.

- (328) FANTHAM (H. B.). *Note on the Occurrence and Distribution of Herpetomonas pediculi.*—*Ann. Trop. Med. & Parasit.* 1912. Oct. Vol. 6. No. 3. B. pp. 403-404.

The author's reason for drawing attention to this parasite is the attention that is being paid to the problems of the transmission of leishmaniasis, spirochaetosis, and trypanosomiasis.

The organism is a natural parasite of the body and head louse, and has no connection with either *Leishmania* or *Trypanosoma*.

The parasite has been observed in England and in India and Tunis.

REPORTS.

- (329) GOLD COAST. *Report of the Veterinary Department for 1911.* [Beal (W. P. B.).]—1912. Accra: Printed at the Government Press.

This Report comprises an account of the clinical work and farriery, cattle trade and statistics, slaughterhouse statistics, tours of inspection, brief descriptions of the contagious and infectious diseases met with, which include anthrax, contagious bovine pleuro-pneumonia, epizootic lymphangitis, filariasis, fowl cholera, piroplasmosis, septicaemia haemorrhagica and trypanosomiasis. In the treatment of the latter disease some success appears to have been obtained with arsenic in various forms combined with different salts of antimony. Eight cures are said to have been obtained out of twenty horses treated. In connection with preventive measures Beal has recommended that horses should be given 20 grains of orsudan on alternate days when passing through fly country. In particular fly belts it is recommended that the horses be smeared over with a preparation of pounded tobacco rubbed down with "Moshi" butter, this salve being lightly applied over the whole body.

Experiments were also made to see whether the administration of arsenical preparations would act as a prophylactic in fly districts. Thirteen horses were taken out on manoeuvres during the months of February, March and April. Three of these died of trypanosomiasis and Beal is informed that this is the lowest rate of mortality that has occurred since horses were taken out on manoeuvres. Another horse was "well drugged with arsenic and smeared" and sent to a noted fly district. The animal worked there for ten days and then returned to Accra. No trypanosomes were discoverable in its blood on its return. The following flies have been identified:—*G. palpalis*, *G. tachinoides*, *G. fusca*, *G. nigro-fusca*, *G. longipalpis* and *G. morsitans*. The report includes a list of fourteen helminths met with, and also a list of five grasses said to be of good feeding value.

(330) BRITISH EAST AFRICA. Report of the Department of Agriculture, Nairobi. 1911-1912. [MS.]

Report of the Veterinary Department.

East Coast Fever.—In the report regarding this disease the following points are dealt with:—The history of the disease in the Protectorate; Immunity; The division of the country into “clean” and “infected” areas, with the limits of each; Measures for dealing with the disease, and the regulations which should be in force to control the movements of cattle within the Protectorate.

Contrary to what is found to be the case in South Africa, it is said that calves are relatively less susceptible than adult animals, and that this fact can be used with advantage, in that young animals can be exposed to infection, and, acquiring immunity, reduce the losses for which the disease is responsible. Acting on the lines suggested by this fact it is held that the disease could be to a great extent controlled by allowing the calves to acquire immunity early in life.

Facts show that the country can be divided into two areas, one clean and one infected. The clean area occupies the central portion of the Protectorate, and is somewhat triangular in shape, the base being directed towards the north. This clean area almost divides the infected area into two. On the west the infected area extends right into Uganda, and on the east to the coast.

The plan of causing calves to become infected is not applicable to the clean area, and the measures for the protection of this area against contamination are directed towards preventing the entry of infected cattle into it.

The following summary of the regulations for the control of movement of cattle within the Protectorate is given:—

- (1) Within clean areas movement should only be made under permit.
- (2) From clean area to clean area through an infected area movement should be by rail only and under permit.
- (3) From a clean area to an infected area: Movement under permit, and the branding of cattle entering highly endemic areas.
- (4) Movement within infected areas by permit, save that cattle from a slightly infected area should not be moved to a highly endemic area unless they are branded.
- (5) Movement from highly endemic areas to clean areas under permit.
- (6) Immune cattle to be moved with permit throughout the Protectorate.
- (7) Cattle used for transport to have passed the immunising test, to have a distinguishing mark, and to be accompanied by certificate.

Rinderpest.—Practically the whole of the Nyanza Province is infected, and the mortality is great. Efforts to suppress the disease by the application of protective serum were to a great extent rendered useless owing to the movement of cattle by the natives. Isolated outbreaks have occurred in other parts and these have been promptly arrested by serum treatment.

Pleuro-pneumonia exists in one district only, and the herds have been confined to an isolated area.

No cases of *anthrax* have been verified.

Anaplasmosis is fairly prevalent, but no serious losses have been reported.

Cases of *trypanosomiasis* in cattle have been observed somewhat frequently on some of the rivers and on Victoria Nyanza, and

among native cattle on the coast, but no cases have been reported from elsewhere.

Coccidiosis of bovines has been occasionally reported, but no serious loss has been recorded.

Horse sickness has been reported in a few isolated cases, but indications are not wanting that during the coming season the disease may be very prevalent.

Epizootic lymphangitis has increased to a large extent, cases having been recorded in practically every settled district. The fact that an infected horse is still capable of work for some time renders control of this disease difficult.

A disease resembling ulcerative lymphangitis is reported from the Uasin Gishu Plateau. This condition is a chronic one extending to several months. The disease is characterised by the appearance of discrete hard crusts under the skin, which may make their appearance all over the body. In time these tend to extend and coalesce, the detachment of the raised crust with the hair involved leaving slowly healing ulcers. There is no appreciable exudate.

The principal diseases of the sheep are briefly dealt with, scab being considered at the greatest length.

Pleuro-pneumonia is prevalent among goats in certain districts, but segregation has prevented the spread.

Sarcoptic scabies has been responsible for the death of a number of goats. The native sheep are also susceptible to this disease, but not to so great an extent as the goat. The wool breeds appear to be immune.

Swine fever in the Protectorate is a very much more severe disease than that known in Europe, practically every pig that becomes infected dying.

Tick fever in dogs has been very prevalent, but the use of trypanblue has been extremely valuable as a curative agent.

Distemper has caused considerable losses at some places.

Report of the Veterinary Pathologist.

In connection with East Coast Fever experiments have been made on the following lines:

1. To prove by the agency of what ticks the transference is effected.
2. To ascertain what immunity is possessed by young calves.
3. To ascertain the value of inoculation by intraperitoneal, intrasplenic, and intrajugular of spleen or other organ pulp.
4. To prove that East Coast Fever in the Protectorate is identical with that occurring in South Africa.
5. Medicinal treatment.

In connection with the first of these it has been proved that *R. appendiculatus* having fed as a larva upon a infected animal will transmit the disease as a nymph, and probably a nymph feeding upon an infected animal will transmit as an adult, but proof of this has not been furnished. So far experiments with *R. caveri* have failed. It appears to be possible that the "yellow backed" tick (*R. pulchellus*) may prove to be capable of transmitting the infection.

Experiments are at present being carried out to ascertain whether calves have a higher degree of resistance than adult animals, and some of the results obtained appear to suggest that such is actually the case.

Experimental inoculations have had variable results, and appear to suggest that material from every animal is not of the same value for inoculation purposes, and that therefore the results depend to a great extent upon the material used. It remains to be ascertained what are the means of recognising good from bad material. Experiments have conclusively proved that the disease occurring in the Protectorate is identical with that observed in South Africa.

No attempts at treatment have influenced the course of the disease. In this connection one experiment with "606" has been made. A spirochaete, probably *S. theileri*, has been encountered both in specimens of blood sent into the laboratory, and in the blood of some of the laboratory animals. In the latter no symptoms were observed, though the parasite is credited with causing anaemia and death by the owners of infected cattle.

Among the affections due to worms mention is made of the occurrence of *Bilharzia crassa* in the portal vein of oxen coming from Uganda, and also of filarial embryos in the peripheral blood.

Among the conditions referable to the presence of trypanosomes in the blood of equines mention is made of one particular parasite which originated from a camel coming from Boran. This parasite is under study in Uganda, and it is interesting to note that it has not been found possible to transmit it by means of the common tsetse fly *G. palpalis*.

Another trypanosome, probably a new species, was encountered in some mules taken to Voi, where they remained a few days only. The symptoms presented by some of the affected animals were comparable to those of horse sickness. The trypanosome was of very low virulence for all the other animals experimented upon.

In connection with the occurrence of horse sickness it is stated that while a few cases of this disease have been observed there is no doubt that many animals show swelling of the head and other symptoms closely simulating those of horse sickness but not caused by this virus. Such cases have been observed in animals affected with trypanosomiasis and nuttaliasis.

A few cases of nuttaliasis have been observed, and the only tick which has so far been proved to be a transmitting agent is *R. evertsi*.

In certain localised parts there appears to be a disease of donkeys characterised by anaemia, wasting, and dropsy in which no piroplasm or trypanosome can be discovered.

Blue tongue in sheep is recorded on various farms, and it is believed that in some cases mild attacks occur which escape notice, but result in great loss of condition, loss of wool, and abortion. A vaccine can be prepared against this disease. Swine fever encountered in British East Africa is far more fatal than that occurring in Europe. Scarcely a pig survives. At post-mortem examination there are found marked inflammation of the stomach, great enlargement of the spleen, petechiae in the kidneys, often

large haemorrhagic areas on the heart, and oedema of the lungs. In the large bowel there may be ulceration. Investigations would appear to indicate that the disease is distinct from that occurring in Europe, one point suggesting this being that the Africa virus is far less resistant to heat, 60° C. destroying it in ten minutes, whereas the European virus resists this temperature for 24 hours. Further investigations suggest that the disease is contracted by the ingestion of infective material, and that the infective material does not retain its virulence for long outside the body.

BOOK REVIEW.

(331) HOARE (E. W.). **A System of Veterinary Medicine.** By various Writers. Edited by E. Wallis Hoare. 1st Edition. Vol. 1. 1327 pages. 1913. London: Ballière, Tindall & Cox. [42s. net.]

This volume is devoted to the microbial diseases, and the portion of it relating to diseases occurring in tropical countries, or of special interest to veterinarians in tropical countries extends to some three hundred pages. Piropiasmosis, trypanosomiasis, leishmaniasis, spirochaetosis, heartwater, blue tongue, horse sickness, epizootic lymphangitis, rabies, Mediterranean fever, and rinderpest are dealt with. As the book is not primarily devoted to the consideration of tropical diseases it is obvious that the accounts of the tropical diseases cannot be exhaustive. The fact that the descriptions have been written for the most part by authors in tropical countries who come into direct contact with the diseases described is of great value. The additions to our knowledge regarding tropical diseases follow each other so rapidly that it is impossible for a book of this nature to be quite up to date at the time of publication, but nevertheless as a general text-book the volume is valuable.

It is to be regretted that the term *Piroplasma parvum* is used in the section dealing with East Coast Fever, although it is pointed out in the introductory paragraphs that *Theileria parva* is the name now generally accepted for the designation of this parasite, and in the same portion of the book there occur a few errors in the spelling of names. The life cycle of *T. parva* is passed over with a bare mention that work has been done on it and that Koch's bodies are specific. The non-inoculability of the disease by means of blood does not appear to have received mention save in the introduction to the subject, where it is briefly referred to. In the section dealing with the coccidiosis of the rabbit it is said that in the process of formation of the sporoblasts the protoplasm of the oocyst divides into two spheres, each of which again divides into two; whereas it is known that the four sporoblasts are formed simultaneously.

It is a matter for regret that the form in which the references are given is not consistent throughout the book. In some portions the references are given in their entirety, while in others the source of information is indicated only by the name of the journal and the year.

The volume includes a complete index.

The general plan followed in the various chapters is good, and the information is well arranged.

RECENT LITERATURE.

[Continued from BULLETIN No. 2, pp. 132-138.]

Babesiasis.

- (332) BOUET (G.) & ROUBAUD (E.). La Piroplasmose (Nuttalliose) de l'Ane en Afrique Occidentale. [Piroplasmosis (Nuttalliasis) of the Donkey in West Africa.]—*Bull. Soc. Path. Exot.*, 1912. Dec. Vol. 5. No. 10, pp. 806-808.
- (333) GILRUTH (J. A.). The Introduction and Spread of the Cattle Tick (*Boophilus annulatus*, var. *Microplus*), and of the Associated Disease Tick Fever (Babesiasis) in Australia.—*Proc. Roy. Soc. Victoria*, 1912. Aug. Vol. 25. (New Series.) Pt. 1, pp. 15-22.

Filariasis.

- (334) LOW (G. C.). The Life of Filarial Embryos outside the Body.—*Jl. Trop. Med. & Hyg.*, 1912. Nov. 15. Vol. 15. No. 23, pp. 338-339.
- (335) MITTER (S. N.). *Filaria immitis* in Calcutta.—*Bull. Soc. Path. Exot.*, 1912. Nov. Vol. 5. No. 9, pp. 731-733.
- (336) SAISAWA. Untersuchungen über Hundefilarien. [Researches on the Dog Filaria.]—*Centralbl. f. Bakt.*, 1. Abt., Orig., 1912. Nov. 9. Vol. 67. Nos. 1/2, pp. 68-75. With 2 plates and 1 text-figure.

Fowl Pest.

- (337) LANDSTEINER (Karl) & BERLINER (Max). Ueber die Kultivierung des Virus der Hühnerpest. [The Cultivation of the Virus of Fowl Pest.]—*Centralbl. f. Bakt.*, 1. Abt., Orig., 1912. Dec. 4. Vol. 67. No. 3, pp. 165-168.
- (338) MROWKA. Das Virus der Hühnerpest ein Globulin. [The Virus of Fowl Pest is a Globulin.]—*Centralbl. f. Bakt.*, 1. Abt., Orig., 1912. Dec. 11. Vol. 67. No. 4, pp. 249-268.

Leishmaniasis.

- (339) BASILE (C.). Sur l'Identité des Leishmanioses et sur leur Mode de Transmission. [The Identity of the Leishmaniasis (in Children and Dogs) and their Methods of Transmission.]—*Bull. Soc. Path. Exot.*, 1912. Dec. Vol. 5. No. 10, pp. 812-814.
- (340) MARZINOWSKY (E. I.). Maladies Voisines de la Malaria en Russie, Kala-azar, Fièvre de Malte, &c. [Diseases related to Malaria, Kala-azar, Malta Fever, &c., in Russia.]—*Bull. Soc. Path. Exot.*, 1912. Dec. Vol. 5. No. 10, pp. 868-876. With 2 plates.

Leprosy.

- (341) LEBOEUF (A.). Dans la Lèpre chez l'Homme, comme chez le Rat, on peut trouver des Bacilles Spécifiques dans les Ganglions superficiels. [Specific Bacilli occur in the superficial Glands in Leprosy in Man as in the Rat.]—*Bull. Soc. Path. Exot.*, 1912. Oct. Vol. 5. No. 8, pp. 569-571.
- (342) MARCHOUX (E.) & SOREL (F.). La Lèpre des Rats. (Second Mémoire.) [Leprosy of the Rat.]—*Ann. Inst. Pasteur*, 1912. Oct. 25. Vol. 26. No. 10, pp. 778-801.

Malaria.

- (343) SINTON (J. A.). Some Attempts at the Cultivation of the Malarial Parasite by Bass's Method.—*Ann. Trop. Med. & Parasit.*, 1912. Oct. 18. Vol. 6. No. 3 B, pp. 371-373.
- (344) THOMSON (J. G.) & McLELLAN (S. W.). The Cultivation of one Generation of Malarial Parasites (*Plasmodium falciparum*) in vitro, by Bass's Method.—*Ann. Trop. Med. & Parasit.*, 1912. Dec. 30. Vol. 6. No. 4, pp. 449-462. With 2 plates and 2 charts.

Plague.

- (345) MARKL. Bakteriologische Diagnose der Rattenpest. [Bacteriological Diagnosis of Rat Plague.]—*Centralbl. f. Bakt.*, 1. Abt., Orig., 1912. Dec. 30. Vol. 67. No. 5, pp. 388-397.

Rabies.

- (346) MANOUÉLIAN (Y.). Etude des Corpuscles de Negri et des Formations Spéciales à la Rage à Virus Fixe. [The Nature of Negri Bodies and Peculiar Alterations found in Rabies caused by the Fixed Virus.]—*Ann. Inst. Pasteur*, 1912. Dec. Vol. 26. No. 12, pp. 973-985. With 2 plates.
- (347) PORAK (R.). Des Altérations fonctionnelles des Glandes Sur-rénales dans la Rage. [Functional Changes in the Suprarenal Bodies in Rabies.]—*Compt. Rend. Soc. Biol.*, 1912. Dec. Vol. 73. No. 35, pp. 601-602.

Rinderpest.

- (348) HARTLEY (P.). Report on the Preparation of Rinderpest Anti-Serum by Means of Diluted Virulent Fluids.—*Indian Civil Veterinary Dept. Memoirs*. No. 3. [Period covered 1910-1911.] pp. 231-241.

Spirochaetosis.

- (349) VON PROWAZEK (S.). Einfluss hämolytischer Stoffe auf Spirochäten (Spirochaetaceae). [The Effect of Haemolytic Substances upon Spirochaetes.]—*Centralbl. f. Bakt.*, 1. Abt., Orig., 1912. Oct. 12. Vol. 66. Nos. 5/6, pp. 424-426.

Theileriosis.

- (350) CARPANO. La Febbre della Costa nella Colonia Eritrea. Note Biologiche e Morfologiche sulla *Theileria parva*. [East Coast Fever in the Colony of Eritrea. Biological and Morphological Notes on *Theileria parva*.]—*Clinica Veterinaria*, 1912. Nos. 19, 20, 21, 22.
- (351) FRANÇA (Carlos). Quelques Considerations sur le Genre *Theileria* et Description d'une Nouvelle Espèce de ce Genre (*Theileria stordii*). [On the Genera *Theileria* and on a New Species of this Genus (*Theileria stordii*).]—*Centralbl. f. Bakt.*, 1. Abt., Orig., 1912. Dec. 4. Vol. 67. No. 3, pp. 171-174.

Trypanosomiasis.

- (352) BATTAGLIA (Mario). Einige anatomo-pathologische Läsionen bei der Nagana (*Trypanosoma brucei*). [Some Anatomo-pathological Lesions in Nagana.]—*Centralbl. f. Bakt.*, 1. Abt., Orig., 1912. Dec. 4. Vol. 67. No. 3, pp. 168-170.
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- (359) ADIE (Helen). Note on the Sex of Mosquito Larvae.—*Ann. Trop. Med. & Parasit.*, 1912. Dec. 30. Vol. 6. No. 4, pp. 463-466. With 1 plate.
- (360) AUSTEN (E. E.). New African Tabanidae. Parts II and III.—*Bull. Entom. Research*, 1912. Nov. Vol. 3. Pt. 3, pp. 329-338; and Dec. Pt. 4, pp. 399-416. With 4 text-figures.
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- (375) ASHWORTH (J. H.) & RETTIE (T.). On a Gregarine-*Steinina rotundata*, n. sp.—present in the Mid-Gut of Bird-Fleas of the Genus *Ceratophyllus*.—*Proc. Roy. Soc.*, 1912. Dec. 17. Vol. B 86. No. B 584, pp. 31-38. With 1 plate.
- (376) FANTHAM (H. B.) & PORTER (Annie). Some Effects of the Occurrence of Myxosporidia in the Gall-Bladder of Fishes.—*Ann. Trop. Med. & Parasit.*, 1912. Dec. 30. Vol. 6. No. 4, pp. 467-481.
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BABESIASIS.

- (385) VREIBURG (A.). Einige Untersuchungen über *Babesia bigemina*. [Some Experiments with *Babesia bigemina*.]—*Zeitschr. f. Infektionskrankh. parasit. Krankheit. u. Hygiene d. Haust.* 1913. Mar. Vol. 13. No. 3-4. pp. 180-186.

Attempts to cultivate Babesia bigemina in vitro.—

Blood was withdrawn from the jugular vein of a calf which was suffering from a relapse of the disease without haemoglobinuria. The blood was carefully defibrinated without frothing and 50 per cent. sterile solution of dextrose added in the proportion of 1 per cent. This mixture was then centrifuged.

After complete centrifugalisation the layer of serum was carefully pipetted off and distributed into tubes, the red corpuscles being then withdrawn by passing the pipette right to the bottom of the tube. The red corpuscles were ejected into the bottoms of the tubes in which the serum had been previously placed. According to BASS and JOHNS, who devised the method for the cultivation of the malaria parasite, red blood corpuscles alone can be obtained by this method, but the present author was unable to get red corpuscles quite free from leucocytes.

Tubes so prepared were incubated at 37° C. and 22° C. respectively.

Subcultures were made on the third day, the blood mixture being prepared in the same way as before save that blood from a healthy animal was used. Blood corpuscles were withdrawn from the primary tubes and added to the fresh tubes, the quantity added being about a fifth of the volume. These were also incubated at 37° C. and 22° C.

After a further three days a third series of tubes was inoculated from the second. Smear preparations were examined from the different tubes at intervals.

In each smear made from the primary cultures incubated at both temperatures two parasites were found on an average, while none were found in control tubes containing simple defibrinated blood. On the sixth and ninth days no parasites were discoverable in the primary cultures.

No parasites could be found in the cultures of the second generation incubated at 37° C. In a preparation made on the third day from a tube incubated at 22° C. six parasites were found, but none were found on the sixth and ninth days.

No parasites were found in preparations from the third generation of cultures.

In a second series of experiments the dextrose was replaced by glucose, the preparation of the tubes and the incubation being the same as before.

In control tubes of defibrinated blood incubated at 37° and 22° C. no parasites were discoverable on the sixth day.

In the tubes of the first generation incubated at 37° C. parasites were just as numerous as in the blood, but there was no multiplication. A few days later all the parasites had disappeared.

Smear preparations from the tubes incubated at 22° C. showed that multiplication had occurred. Parasites, most of which were of the amoeboid type, were found both intra- and extra-corpuseular in position, and a very small number of double forms was encountered. A few days later some coccus-like bodies were found both within and external to the blood corpuscles. After twelve days no parasites could be found.

The tubes of the second generation showed about six parasites per smear on the third day, and after about the thirteenth day none could be found.

There was no multiplication in the tubes of the third generation incubated at 37° C.

In a preparation made from a tube incubated at 22° C. a number of rounded bodies measuring 1 to 2 microns in diameter were found, some within and some external to the blood corpuscles. These could not be found ten days later.

A further series of tubes incubated at 22° C. failed to show any multiplication of the parasites.

Attempts to cultivate the parasite in the condensation water of blood agar and of dextrose blood agar failed.

The small variety of Babesia bigemina.—

After giving a brief abstract of McFADYEAN and STOCKMAN's description of *Babesia divergens* the author states that during very frequent examinations of the blood of cattle suffering from haemoglobinuria in Holland he has only encountered small parasites. The majority of the organisms are either irregularly rounded or slender. Pear-shaped parasites are not found. The slender forms have a bluntly rounded end and a pointed end, and they vary in length from 1.5 to 2, and exceptionally 2.5 microns in length. The rounded forms have a maximum diameter of 1.5 microns.

Single parasites are more frequently observed in the blood corpuscles than double ones.

In the elongated forms there is a somewhat large mass of chromatin generally disposed at the blunt end, and in the rounded forms the amount of cytoplasm is usually very small.

Details are given of the microscopic examination of the blood of three infected animals and from these the following facts may be gathered:—

The percentage of infected corpuscles may be as high as 40. The rounded forms outnumber the slender forms. Very few parasites are to be found free in the plasma. A greater proportion of the parasites occur singly in the corpuscles, and of these about 50 per cent. are centrally and 50 per cent. peripherally placed. The percentage of single parasites lying at the rim of the corpuscles was 7 in one case and 51 in another. The percentage of double parasites varied from 16 to 30, and these in the great majority of cases did not lie towards the periphery of the corpuscles. In one instance 50 per cent. of these showed marked divergence, but in another it was observed in 3 per cent. only.

In a small proportion of cases three or even four parasites were encountered in a single corpuscle.

Animals infected experimentally with this parasite showed very slight reactions, in some instances there was only slight fever without parasites being discoverable in the blood.

The author concludes that the parasite which occurs in Holland resembles *B. divergens* save that the divergence of the double parasites is not usually observed and that the parasites do not show a special tendency to take up a position at the rim of the corpuscles. He thinks that the name *B. divergens* is not applicable to the parasite and that the organism is a small form of *B. bigemina* such as has been described as occurring in Germany and North Africa.

No cross-immunity experiments have been done, but the author suggests that should these indicate non-identity the name "*Babesia bovis*" should be given to the small parasite.

(386) DSCHUNKOWSKY (E.) & LEUS (T.). *Nuttallia* und *Piroplasma* bei der Piroplasmose der Einhufer in Transkaukasien. [*Nuttallia* and *Piroplasma* in Piroplasmosis of solipeds in Transcaucasia.] — *Parasitology*. 1913. Jan. Vol. 5. No. 4. pp. 289-302. With 2 plates.

In this paper all the cases of piroplasmosis that have been observed by the authors in solipeds are summarised, and the conclusion arrived at supports the view that two species of parasite are concerned—*Nuttallia* and *Piroplasma*.

The first case observed occurred in an imported horse which died as a result of the infection. Examination of the blood in this case showed that about 1 per cent. of the red cells contained parasites. The great majority of these were rounded in shape, but oval, pear-shaped, and amoeboid forms were seen. The authors think that the African parasite, *Nuttallia equi*, is differentiated from that observed in the first case in Transcaucasia by the fact that in the former amoeboid forms are the most commonly observed type of parasite, while the rounded parasites are far fewer in number. A further point of difference is the frequent occurrence of cross-forms in *Nuttallia*, this being the manner in which multiplication occurs in this species.

In mature parasites the chromatin mass is generally single and rounded in shape. It is practically impossible to make out a blepharoplast, but in some parasites the chromatin is present in the form of a longer or shorter curved rod in which darkly stained granules can be observed.

The cytoplasm stains blue, and has a reticulate structure. It appears to be more condensed towards the periphery of the parasite.

The chromatin is nearly always surrounded by a clear zone.

The authors mention a striking character possessed by the parasites studied by them, namely, a marked variation in size. The rounded forms varied from 0.8 to 2.8 microns in diameter, and the pear-shaped forms from 1.5 to 4 microns in length, by 1.0 to 1.5 microns in width.

They are of the opinion that the smallest parasites do not represent young forms but mature organisms, and, consequently, they think that they are not dealing with a single organism, but with two separate varieties—a large and a small. They are not at present in a position to say which parasites these represent.

The possibility is suggested that in Transcaucasia there are three distinct types of parasite—a large and a small *Nuttallia*, and *Piroplasma caballi*.

The cases of the disease observed in 1906 and 1907 were all due to parasites of the genus *Nuttallia* save one which was caused by *Piroplasma*.

In some cases of *Nuttallia* infection the percentage of infected corpuscles rose as high as 30 to 40.

At the post-mortem of the horse in which there was a mixed infection the only lesions found were great enlargement of the spleen (up to 5 or 6 times) and yellow discoloration of the mucous and serous membranes. There were no abnormalities observable in the blood save the presence of parasites. The disease ran an acute course.

The piroplasms presented a typical pear-shaped form resembling the large parasites seen in bovine piroplasmosis. In a minority of cases two parasites were present in the same corpuscle, but more than two were not observed. The parasites were joined together by their thin ends, and in many cases there was a small round granule of chromatin at the point of junction.

The large parasites varied from 2.4 to 3.7 microns in length and from 1.0 to 1.5 in width. In practically all the pear forms a nucleus and blepharoplast could be made out.

Some of the pear-shaped or oval parasites showed bud-like processes which appeared to suggest that the parasites were dividing. There were two bud-like processes in every instance and in the great majority of cases they were placed on one side of the parasite. After their liberation from the parent cell these processes appeared to form twin pear-shaped parasites which afterwards separated from each other.

In some of the pear-shaped organisms the chromatin breaks up into a number of fragments which pass either to the periphery of the parasite or remain scattered through the cytoplasm.

The amoeboid forms which are always observed are distinguishable from the budding forms already described. They have

a very irregular outline and possess pseudopodia which are either pointed or thickened at the ends.

The forms already described have been observed in blood taken during life.

In the blood of an animal which had been dead five hours only round parasites were seen which had a shrunken appearance.

With regard to the pear-shaped parasites of the two genera, the authors make the following statements:—

In *Nuttallia* pear forms are far more scanty than round forms. In *Piroplasma* the pear form is by far the most common and characteristic. In *Nuttallia* pear forms in most cases occur singly in the corpuscles, and when two are present in a single cell they are not united by their thin ends. In *Piroplasma* double pear forms commonly occur and these are generally joined together. In *Nuttallia* the nucleus and the blepharoplast are practically always together, while in *Piroplasma* they are quite easily distinguishable from each other. Further, in the former the nucleus is practically always round while in the latter it is often elongated or fragmented. The structure of *Piroplasma* appears to be more complicated than that of *Nuttallia* owing to the presence of vacuoles in the cytoplasm and to the fragmentation of the chromatin.

In all, twelve cases of the disease have been observed, and in only one of these was *Piroplasma* found and then in conjunction with *Nuttallia*.

In one instance infection with *Nuttallia* was observed in a mule. Two donkeys and one horse were inoculated with large doses (500, 100, and 800 c.c.) of defibrinated blood. In one animal only, the donkey which received 100 c.c., was there any reaction, and parasites were found in the blood on the thirteenth day.

In order to gain some idea as to the prevalence of the disease among donkeys, 18 animals which were unfit for work were examined, and eight were found to be infected. Eleven of the donkeys died in the course of two months, and four were found to have parasites in their blood. At the post-mortem examination no abnormalities save the lesions of anaemia were discovered. There was no swelling of the spleen.

Four of the infected donkeys survived.

The authors think that the parasite found in the donkeys is possibly a definite species of *Nuttallia*, but they do not make a definite pronouncement on the point.

- (387) VON RÁTZ (S.). Ueber die Piroplasmose der Schafe. [Piroplasmosis in the Sheep.]—*Centralbl. f. Bakt.* 1 Abt., Orig. 1913. Mar. 1. Vol. 68. No. 2. pp. 194-200. With 2 text-figures.

After reviewing the literature regarding the occurrence of piroplasmosis in the sheep, the author refers to an outbreak of disease in sheep in Hungary in 1909, in which piroplasms were discoverable in the blood. In view of the facts that the lesions found on post-mortem were not indicative of piroplasmosis, and

that inoculation experiments failed no great importance was attached to the discovery.

In the autumn of 1911 Balás found the following lesions in a sheep which had been slaughtered:—

Numerous punctiform haemorrhages in the subcutaneous connective tissue. Anaemia of all the organs and a laked condition of the blood. A small number of haemorrhages in the serous membranes, and slight swelling of the spleen. Catarrh of the mucous membrane of the large and small intestines associated with punctiform haemorrhages, particularly in the region of the pylorus. Numerous haemorrhagic infarcts of various sizes in the lungs. Acute swelling of the lymphatic glands.

The spleen was forwarded for examination, and in smears stained by the May-Grunwald method piroplasms were discovered in considerable numbers.

There were practically no abnormalities to be observed in the red corpuscles, and they took the stain fairly well. Only those cells which contained from 1 to 3 large parasites showed any enlargement.

The parasites were very variable in size and shape. They measured from 9·8 to 2·0 microns in diameter, and rounded, oval, rod-shaped, and ring-forms were seen. In the majority of cases the parasites were surrounded by a lighter zone. Save in a few instances they were placed peripherally in the infected corpuscles. The smallest of the parasites resembled cocci, but they showed in their centre a minute nucleus which stained of a bluish red tint. The larger round parasites presented a similar appearance, but their nuclei were proportionately larger. In some cases they appeared to be dividing longitudinally, and in each portion there was a piece of chromatin. When pairs of small parasites occurred they presented an appearance suggestive of diplococci.

Oval forms were not uncommon, and in these the nucleus was situated at or near one of the poles. In the majority of cases these parasites occurred singly in corpuscles, but in some cases the same cell contained either mono- or diplococcus forms in addition. Some of the round or oval forms possessed irregular pseudopodia. Short thick rod-like forms occurred in small numbers and in these there was a piece of chromatin at each end. Curved rod forms were observed applied to the outer surface of red corpuscles.

The most characteristic forms were the pear-shaped organisms which occurred singly or in pairs. In the centre of these larger parasites there was a distinct nucleus, and in many of them a small fragment of chromatin, possibly representing a blepharoplast, could be made out at one end. When two pear-shaped parasites occurred in a single corpuscle they were as a rule smaller than the single ones. The relative positions occupied by the two parasites varied greatly. In some cases they were nearly parallel to each other and in others they formed a wide angle or even a straight line.

A few ring forms were observed. In these the central part appeared vacuolar, and a rounded or elongated nucleus could be seen in the cytoplasmic rim.

In a few large corpuscles, which did not stain intensely, rosette forms of reddish granules could be seen. In many of these no cytoplasm could be distinguished.

Parasites at different stages of development could be found in a single corpuscle, and very few parasites were seen free in the plasma.

In the second outbreak of the disease a number of lambs were attacked, the ewes and the very young lambs escaping infection.

The lesions found at the post-mortem of one of these animals did not suggest piroplasmosis, but the parasite was discovered in the blood. The principal lesions were: Enlargement of the lymphatic glands, parenchymatous degeneration of the kidneys, liver, and cardiac muscle, sero-fibrinous pericarditis and haemorrhages under the epicardium.

The parasites present in the blood exactly resembled those seen in the previous case, save that the rounded and amoeboid forms were the most numerous.

In some of the pear-shaped parasites indications of division could be observed.

Subsequently the author found piroplasms in smears from three spleens which were forwarded for examination.

It would appear that two forms of the disease occur, an acute and fatal form, and a mild and chronic or latent form. In many cases the lesions are so little characteristic that only examination of the blood allows a diagnosis to be made.

(388) KNUTH & RICHTERS. *Über die Vermehrung von Piroplasma canis in vitro.* [The Multiplication of *P. canis* in vitro.]—*Berl. Tierärztl. Wochenschr.* 1913. Mar. 20. Vol. 29. No. 12. pp. 211-212.

The authors have not strictly followed the technique of BASS as did ZIEMANN in his successful cultivation of *P. canis*.

In the original tube blood containing a few parasites was mixed with one per cent. dextrose solution in the proportion of two to one. The mixture was defibrinated and freed from leucocytes by centrifugalisation. The tube was then incubated at 40° C. About $\frac{1}{2}$ - $\frac{1}{3}$ cc. of this mixture was then added to a tube containing 8 cc. of normal dog blood to which 4 cc. of one per cent. dextrose solution had been added. This mixture was also defibrinated, and centrifuged for the removal of the leucocytes. In both of these tubes there was a multiplication of the parasites in 18 to 20 hours.

In subsequent experiments the serum was not removed and a two or even three per cent. solution of dextrose was substituted for the one per cent. solution originally used. Some of the tubes were incubated at 40-41° C., some at 37.5° C., and some were left at room temperature.

The best results were obtained in those tubes which contained a mixture of defibrinated blood and a two per cent. solution of dextrose and which were left at room temperature. In these tubes there was a distinct multiplication of the parasites in eight days.

THEILERIASIS.

- (389) CARPANO (M.). Su di un Piroplasma del tipo Parvum (genus *Theileria*) riscontrato nella Gazzella in Eritrea. Nota di priorità. [A Piroplasm of the Parvum type (genus *Theileria*) in a Gazelle in Eritrea.] — *La Clinica Veterinaria*. 1913. Mar. 30. Vol. 36. No. 6. pp. 254-256.

In 1909 while travelling in Eritrea the author saw a gazelle infected with a piroplasm of the parvum type. The animal was in good condition, and all its organs including the spleen were normal in appearance. The parasite was not very numerous present in the blood, about two infected corpuscles being found in each field of the microscope (1/15 inch objective and ocular 6).

In the great majority of cases each infected corpuscle contained a single parasite only, and corpuscles containing more than two were exceedingly rare.

The following forms were observed:—

Small rounded forms which appeared to be composed almost entirely of chromatin.

Round or oval ring forms. These stained blue, and showed at one side two masses of chromatin, which were either rounded, curved, or ring-shaped.

Pear or club-shaped forms. These, like the preceding, measured from one to one and a half microns in diameter. The masses of chromatin in the majority of instances were placed at the thinner end of the parasite.

Comma forms. In these the protoplasm was coloured throughout and the mass of chromatin was either round or elongated.

Bacillary forms. These measured from 2 to 3 microns in length. In these also the chromatin was either rounded or elongated in shape.

The comma forms and the bacillary forms were the most numerous, then the oval and round forms. In cases in which four parasites were present in a single corpuscle they were arranged in the form of a cross.

TRYPANOSOMIASIS.

- (390) BRUCE (D.), HARVEY (D.), HAMERTON (A. E.), DAVEY (J. B.), & Lady BRUCE. The Trypanosomes found in the Blood of Wild Animals Living in the Sleeping-Sickness Area, Nyasaland. — *Proc. Roy. Soc.* 1913. Apr. 7. Series B. Vol. 86. No. B 587. pp. 269-277.

In this paper the greater part of the information given is expressed in tabular form. The first table gives a list of 180 animals shot in the fly area on the shores of Lake Nyasa whose blood was examined for trypanosomes. In the great majority of cases the examination included the preparation of thick and thin smears, and the inoculation of goats, monkeys, and dogs. Of these 180 animals 57 or 31·7 per cent. were found to harbour pathogenic trypanosomes.

The second table gives the species of trypanosomes found in the various infected animals. With regard to *T. rhodesiense* the following paragraphs may be quoted verbatim.

"In a previous paper the trypanosome causing human trypanosome disease in Nyasaland was called *Trypanosoma rhodesiense*, on account of the presence of posterior-nuclear forms. This trypanosome agreed in all other respects with *Trypanosoma brucei*, the common trypanosome of wild animals in South Africa, and the cause of tsetse-fly disease, or Nagana. In order to compare the two species of trypanosomes more closely, the Commission procured, by the kindness of Dr. A. Theiler, C.M.G., Pretoria, a strain of nagana from the same spot in Zululand where it was first discovered in 1894. Much to the surprise of the Commission it was found that *T. brucei* has quite as large a proportion of posterior-nuclear forms as *T. rhodesiense*, and that the blunt-ended character is common to both species. The Commission is therefore driven to the conclusion that *T. rhodesiense* is neither more nor less than *T. brucei*, and that the human trypanosome disease of Nyasaland is Nagana. To this it may be objected that Nagana has never been known to attack human beings. This has probably been due to faulty diagnosis, cases in man being returned as malaria."

It would appear from the tables given that among the 180 animals examined *T. rhodesiense* (vel *brucei*) was encountered 14 times, *T. pecorum* 26 times, *T. simiae* 3 times, *T. caprae* 20 times, and *T. ingens*, which is not considered to be pathogenic, 3 times. Although the numbers are too small to be taken literally it is shown that waterbuck, hartebeeste, reedbuck, and duiker are most dangerous as far as man is concerned; eland, koodoo, bushbuck, and buffalo to cattle, goats, and sheep; and that the warthog is the only animal that harbours *T. simiae*, "the lightning destroyer of the domestic pig."

No pathogenic trypanosome has as yet been discovered in the blood of animals living in fly-free areas.

(391) BRUCE (D.), HARVEY (D.), HAMERTON (A. E.), & Lady BRUCE. Morphology of Various Strains of the Trypanosome causing Disease in Man in Nyasaland. I.—The Human Strain. —*Proc. Roy. Soc.* 1913. Apr. 7. Series B. Vol. 86. No. B 587. pp. 285-302.

In this paper the morphology of four strains of the human trypanosome is described in great detail, the results of the measurements made being expressed in a number of tables and curves. In the tables are incorporated the measurements made of a fifth strain of the trypanosome (Strain I, Mkanyanga) which have already been published. (See this *Bulletin*, Vol. 1, No. 2, Ref. No. 204, p. 134.)

Comparison is instituted between the measurements and the curves obtained when the trypanosomes measured were in the blood of various animals (goat, monkey, dog, rat and sheep) and when the measurements were made in a systematic manner on consecutive days of the trypanosomes present in the blood of a single rat.

In the blood of various animals the average length of the five different strains varied from 22·2 to 26·1 microns with an average of 23·5, and in the blood of rats the lengths varied from 22·5 to 26·4, the strains falling nearly, but not quite, in the same order.

In dealing with one of the strains four curves are given. These curves were constructed as follows:—(1) upon 1,500 trypanosomes from various animals, (2) upon 1,000 parasites taken at random, (3) upon 500 trypanosomes taken from a rat on nine consecutive days, (4) as (3), but from a different rat. When these curves are compared it is seen that curves (1) and (2) differ considerably from each other and from (3) and (4) which are very much alike.

To quote the paper, "If curves made in this way from different strains of one species of trypanosome showed the same degree of similarity, this method would certainly be useful for purposes of classification. But, as we have seen, the curve of Strain II has no resemblance to that of Strain I (previous publication), and it will be found that each human strain of this species of trypanosome differs, more or less, when subjected to this method of measurement."

In comparing the curves constructed upon the whole of the five strains the authors express themselves as follows:—

"It must be confessed that, on comparing the five curves one with another, they do not give as much assistance in classifying this species of trypanosome as was hoped. Curves I and III are alike, and coincide with that prepared by Dr. Stephens from the case of Armstrong in Liverpool, whereas Curves II, IV, and V. approach more to the type described by Kinghorn and Yorke from the Luangwa Valley."

In a table showing the percentage of posterior-nuclear forms in the blood of rats inoculated with all the five strains it is seen that it varies from 3·3 to 34·1, with an average of 17·8.

"It is to be noted that in the human strain the percentage of posterior-nuclear forms varies greatly, although the method of enumeration is the same in each case. This presence of posterior-nuclear forms would have been accepted a few months ago as sufficient proof that the species dealt with was *T. rhodesiense*. Since then the posterior nuclear forms have been reported as occurring in *T. brucei* from Egypt, Uganda and Zululand. In a strain lately obtained by Theiler from the same spot in Zululand where this species was originally discovered in 1894, this percentage rose to the highest yet recorded."

Among the conclusions drawn is the following:—

Evidence is accumulating that *T. rhodesiense* and *T. brucei* (Plimmer and Bradford) are identical.

(392) BRUCE (D.), HARVEY (D.), HAMERTON (A. E.), DAVEY (J. B.), & Lady BRUCE. *Trypanosome Diseases of Domestic Animals in Nyasaland*. II.—*Trypanosoma caprae* (Kleine).—*Proc. Roy. Soc.* 1913. Apr. 7. Series B. Vol. 86. No. B 587. pp. 278-284. With 1 plate.

This trypanosome belongs to a group which comprises *T. uniforme*, *T. vivax*, and *T. caprae*. They are all characterised by their extreme motility, clear cell contents, large round

terminal micronucleus, and by the fact that they are not pathogenic for the small laboratory animals. They all develop in the proboscis of flies and not in the alimentary canal. *T. caprae* is transmitted by *G. morsitans*, and the others by *G. palpalis*.

The description of *T. vivax* in the unstained state can, it is said, be equally applied to *T. caprae*.

In the preparation of stained smears the procedure adopted was fixation with osmic acid and staining with Giemsa.

From measurements of the trypanosome in the ox, sheep, goat, and waterbuck the following figures were arrived at:—

| Species of Animals. | Number of Trypanosomes measured. | Average Length. | Maximum Length. | Minimum Length. |
|---------------------|----------------------------------|-----------------|-----------------|-----------------|
| Ox | 40 | 25·7 | 32·0 | 18·0 |
| Goat | 260 | 25·3 | 31·0 | 20·0 |
| Sheep | 180 | 25·6 | 32·0 | 21·0 |
| Waterbuck | 20 | 26·8 | 29·0 | 25·0 |

Total number of trypanosomes measured 500.

A curve constructed upon the measurements of these 500 individuals shows that the organism is monomorphic, the greatest number of trypanosomes (19 per cent.) measuring 25 microns in length.

Measured across its broadest part *T. caprae* measures on an average 3 microns (maximum 4·25, minimum 1·75).

T. caprae differs from *T. vivax* in that it is heavier built and of a more clumsy appearance. The posterior half is swollen, and its end bluntly angular or rounded.

The cell contents have a delicate alveolar structure and are free from granules or vacuoles.

The nucleus is oval and compact and lies about the middle of the body.

The micronucleus is large and rounded and as a rule lies close to the posterior extremity.

The undulating membrane is more developed and is thrown into bolder folds than in *T. vivax*.

There is a well-marked free flagellum which averages 6·5 microns in length.

It would appear from the inoculation of two oxen that the trypanosome is not responsible for serious disease in that animal, as trypanosomes appeared in the blood, in small numbers and for a short time, two months after inoculation, and then disappeared.

In the sheep and the goat the disease set up by this parasite runs a fairly fatal course. Of 36 goats 15 were infected by wild *G. morsitans* and died, the period of infection being from 53-59 days. Four that were inoculated with infective blood died, on an average, in 57 days. The remaining 17 were alive at the time of writing, after intervals of from 61 to 262 days.

Of four sheep inoculated, three died in from 36 to 221 days, the fourth surviving (245 days).

Monkeys, dogs, rats, and guinea-pigs failed to become infected.

The carrier of the trypanosome is *G. morsitans*, and in experiments made to ascertain with what trypanosomes the wild flies are naturally infected *T. caprae* was found in 61 per cent.

The development of the trypanosome in the fly is left for a further communication, but it is stated that it is restricted to the proboscis and runs a course of from 16 to 20 days.

With regard to the reservoir of the trypanosome, it has been found in 10.5 per cent. of 180 specimens of wild game.

(393) MACFIE (J. W. Scott). **Trypanosomiasis of Domestic Animals in Northern Nigeria.**—*Ann. Trop. Med. & Parasit.* 1913. Mar. 31. Vol. 7. No. 1. pp. 1-26. With 3 plates.

During the greater part of 1912 the author was stationed in Ilorin, a province which is not considered unsuitable for horses save in one division, but within a period of eight months ten out of 15 horses in the possession of Europeans contracted trypanosomiasis and six died. In an attempt to introduce animal transport into the province ten donkeys were employed for the purpose between Ilorin town and Agugi, about 30 miles to the east. All were dead of trypanosomiasis within three months. At Zungeru in 1911 25 horses were under treatment, and of these 40 per cent. died or were destroyed. At Lokoja the disease is even more serious and the percentage of dead or incapacitated horses is placed at 75. It is a matter of impossibility to estimate the losses sustained by the natives.

Cattle, sheep, goats, and dogs contract trypanosomiasis, and diseased cattle are killed and sold for food. Ilorin is situated at the point where the main routes from Kano and Sokoto converge and the majority of the animals slaughtered for the market are affected with trypanosomiasis. Examination of 35 animals killed for the market showed that two were infected with *T. brucei*, 30 with *T. vivax*, two with *T. pecorum* or *nanum*, and in one case there was double infection.

In 1911 a list of identifications was published of 15 cases of trypanosomiasis in horses collected in Northern Nigeria. The author has now added a number of fresh cases, both in horses and in other domestic species, bringing the total number up to 86.

The following table gives an analysis of the results of the examinations:—

| Host. | <i>T. brucei</i> . | <i>T. vivax</i> . | <i>T. nanum</i> or <i>pecorum</i> . | <i>T. theileri</i> . | Double infection. |
|------------|--------------------|-------------------|--|----------------------|----------------------|
| Horse ... | 14 | 18 | 8 | — | 3 |
| Donkey ... | 2 | 2 | — | — | — |
| Cattle— | | | | | |
| Fulani ... | 1 | 18 | 1 | — | 1 |
| Dwarf ... | — | 2 | — | 1 | — |
| Sheep ... | 1 | 8 | 1 | — | — |
| Goat ... | — | 4 | — | — | — |
| Dog ... | — | 1 | — | — | — |
| Totals ... | 18 | 53 | 10 | 1 | 4 |

As no systematic experimental inoculations could be carried out the parasites had to be identified by their morphological characters only.

The *T. brucei* referred to is, according to the author, in all probability the trypanosome for which STEPHENS and BLACKLOCK have suggested the name *T. ugandae*. In four cases (three horses and one donkey) of infection with *T. brucei* posterior nuclear forms were detected. In rats and guinea-pigs inoculated from these cases they appeared at certain stages of the disease in fairly large numbers. In a number of films the long forms were observed to have their posterior extremities blunted and almost rectangular. Great variations were observed from day to day in the relative percentages of the long and short forms, and a table is given showing the percentages of the different forms occurring in the blood of a horse, and of a rat and guinea-pig inoculated from it. The parasites are classed as "long forms with free flagellum" and "stumpy and intermediate forms" no measurements having apparently been taken.

It would appear that the disease in equidae is invariably fatal and two forms are clinically recognisable. Acute cases terminate fatally in from two to four weeks, but in chronic cases the disease runs a course of some months. It was noticed that all the cases in which posterior nuclear forms were observed were of the acute type.

T. vivax.—A trypanosome of this type was the one most commonly observed, for, as shown by the table, it was encountered in 56 of the animals examined (53 pure infections and three mixed). In view of the fact that it was found in the dog it was in all probability the *T. vivax* (Ziemann) and not the allied *T. cazalboui* as the latter is not pathogenic for the dog.

The disease caused by this parasite in horses would appear to be mild, since 14 out of 15 infected horses recovered, and in the fatal case the infection was complicated by a simultaneous infection with *T. brucei*. It is difficult to gauge the mortality of the infection in sheep and cattle as the animals in which the trypanosome was seen were slaughtered.

Short trypanosomes of the *nanum* or *pecorum* type were observed in 13 cases, of which ten were horses.

Trypanosomiasis is most common during the rainy season when the tsetse flies are most widely distributed over the country.

The province of Ilorin is peculiar in that whilst *G. palpalis* and *G. tachinoides* are distributed all over it, tsetse flies of the *morsitans* group—*G. submorsitans* and *G. longipalpis*—are restricted to the eastern division.

It is possible that in the native towns and in the European stations flies of the genus *Stomoxys* may play a part in the transmission of trypanosomiasis. One horse which had not been within two miles of any place known to be a fly district for five weeks previous to the onset of symptoms had been tormented by *Stomoxys* flies which were exceedingly common at the time. Both *S. nigra* and *S. calcitrans* have been captured at Ilorin.

Arsenic and perchloride of mercury have been administered both as curative and prophylactic agents but without success.

"No immunity follows an attack of trypanosomiasis, reinfections with the same or a different species of trypanosome being met with."

With regard to the dwarf cattle a paper by Fox is quoted as follows:—

"That a certain breed of cattle found in pagan districts possess a high degree of natural immunity in that they may harbour the trypanosome in the blood and yet keep in good condition and show no signs of the disease, nor do they die from the infection so long as their environments are favourable. These environments are a free life, with ample food, especially plenty of green grass. Confinement, poor feeding, and hard exercise, tend to make the disease manifest itself clinically.

"That such domesticated cattle may act as a reservoir of infection since the blood may prove infective at such times when clinical symptoms manifest themselves, although the trypanosome may not be found on making a microscopical examination."

From another part of the same paper it is quoted that all the trypanosomes met with were of the *T. brucei* type.

Details are given of two experiments carried out on these dwarf cattle, and the results appear to indicate that they, like horses, possess an immunity to *T. vivax*. The immunity however does not appear to be serviceable against *T. brucei*.

A tabular synopsis of the cases of trypanosomiasis in domesticated animals collected in Northern Nigeria is appended. •

(394) BALFOUR (A.). **Animal Trypanosomiasis in the Lado (Western Mongalla) and Notes on Tsetse Fly Traps and on an Alleged Immune Breed of Cattle in Southern Kordofan.**—*Ann. Trop. Med. & Parasit.* 1913. Mar. 31. Vol. 7. No. 1. pp. 113-120. With 2 plates.

The author found trypanosomes in smears of blood taken from transport bulls working on the road running from Rejaf on the Nile to Aba on the Congo frontier. Measurements were taken of 140 specimens and from the results obtained it would appear that the parasite agrees very closely with *T. nanum* occurring in the Sudan. The maximum length of the trypanosome was 17 microns, the minimum 9·8, and the average length 12·6. The parasites varied in breadth from 1 to 2 microns. The infection in none of the three cases was heavy, but the author is informed that it is proving fatal to the cattle. A dog was inoculated with blood containing the trypanosome and parasites appeared in its blood fourteen days after inoculation and then disappeared, and were not found again. The dog remained in good condition. A number of rats which were inoculated died within 2 days from some unknown cause.

If the infection has been derived along the road in Western Mongalla the vector is in all probability *G. palpalis*, as *G. morsitans* is not found in this district, and the only other biting flies in evidence are *Haematopota*. It is, however, probable that the cattle were infected before going to Western Mongalla, and that some species of *Tabanus*, *Chrysops*, or *Stomoxys* is the active agent in transmitting this form of animal trypanosomiasis in the Southern Sudan.

One donkey was found to be infected with trypanosomiasis at Yei, and from an examination of the parasites contained in a single smear it would appear to be not unlikely that the organism was *T. congolense*.

In mules at Yei a trypanosome of the *brucei* or *gambiense* type was found, but gerbils inoculated with blood containing the parasite failed to become infected.

A hundred trypanosomes were drawn and measured and the following results obtained: Minimum length 10.5, Maximum 28, Average 17.6. The trypanosome appeared to be dimorphic, short stout forms without free flagellum and long slender forms with a free flagellum occurring. The short forms measured from 2.5 to 3 microns in breadth and the slender forms 1.2 to 2 microns. In the majority of the short forms there was an unstained area around the blepharoplast, and this was also seen in some of the slender forms.

The breed of small animals said to be immune to trypanosomiasis comes from Southern Kordofan, and it is the only breed that can live in the Koalib district, where *G. morsitans* abounds and conveys an infection which the author believes to be *T. brucei*.

(395) DUKE (H. L.). Some Observations on *Trypanosoma pecorum* (Bruce) and *T. uniforme* (Bruce).—*Proc. Roy. Soc.* 1912. Oct. 11. Series B. Vol. 85. No. B 582. pp. 554-561.

Trypanosoma pecorum.

This trypanosome, according to Bruce and his collaborators, is the cause of a rapidly fatal disease in cattle and domestic animals generally. It appears to be widely distributed throughout Uganda, but nothing definite is known regarding its true carrier in nature. The disease is met with in places where tsetse are unknown and *G. palpalis* would appear from experiments to be at the most a facultative host. Nothing is known regarding a natural reservoir for this trypanosome.

The experiments recorded in the first portion of the paper were carried out with the object of ascertaining whether *T. pecorum* is pathogenic for antelope, and to test whether these animals can act as a reservoir for the trypanosome.

In an experiment, the details of which are tabulated, flies were fed on seven occasions on a bushbuck which had been infected with *T. pecorum* by inoculation from a monkey and from whose circulation trypanosomes disappeared after having been present for some days. These flies were then fed upon clean monkeys, and in only one instance was the result positive. Unfortunately in this instance there was a possibility that the monkey acquired the infection from a source other than the flies. On the occasion of one particular batch of flies having been fed upon the monkey none of the flies were found to contain flagellates, and it was thought that the experiment would of necessity be negative. This monkey was inoculated with blood from two out of three bushbuck shot, a goat being inoculated with mixed blood from all three at the same time. No trypanosomes were found in any of the smears made from these three buck, and the goat did not become infected,

nevertheless there is just the possibility that the monkey acquired the infection from one of the buck and not from the flies.

On dissecting the batch of flies which fed upon the monkey which became infected only one (out of 105) was found to contain flagellates.

A table is given showing the results obtained from the dissection of fifteen infected flies found in the batches used in the experiments with the bushbuck and in another series of experiments in which the infective feeds were obtained on a number of different animals. In the case of three of these flies observations were made upon the sucking stomach and in one only, the one from the batch which in all probability infected the monkey, there were found large numbers of flagellates, and it is suggested that may have a definite developmental significance in *T. pecorum*.

In only three of the flies were the salivary glands obtained, and all were negative. In this respect *T. pecorum* resembles *T. nanum*.

It may also be gathered from the table given that no proboscis infections were obtained before the 76th day of an experiment. Although four flies were obtained showing a good infection of the proboscis none of these were capable of infecting a monkey.

Possibly the flagellates present in the sucking stomach of the one fly were responsible for the infection of the monkey upon which it fed, or there may have been a proboscis infection which had disappeared before the fly was dissected.

It may be further gathered from the table that the bushbuck which was the source of the infection was still capable of infecting laboratory-bred *G. palpalis* three months after its original infection with *T. pecorum*.

Ten months after the original infection of the buck its blood was used in quantities of $2\frac{1}{2}$ to 6 cc. for the inoculation of two monkeys and a calf. The calf, which received 3.5 cc., became infected.

The possibility of error is excluded from this experiment by the following facts:—

1. Since September, 1910, there had been no case of spontaneous infection with this trypanosome among laboratory cattle.

2. The cattle in the neighbourhood of the hill were apparently free from the disease.

3. A calf about the same age as the one used in the experiment had been under observation for some months and had never developed trypanosomes. The two animals had never been allowed to leave the hill top, and had always been stalled together.

The buck that was the source of the infection had not up to the time of writing shown any evidence of having been harmed by the infection. The calf which was successfully infected from it died in 51 days.

The long sojourn of the trypanosome in the bushbuck did not render it more suited to development in *G. palpalis*.

Trypanosoma uniforme.

This organism appears to be the most common antelope trypanosome in the Mpumu neighbourhood. According to Bruce it is responsible for a rapidly fatal disease in domestic ruminants, death occurring in laboratory infected goats in 29 days.

The present author's experience leads him to form a contrary opinion. During a period of 20 months only one animal has died of the infection.

A characteristic feature of *T. uniforme*, which is especially marked in goats, is the manner in which, after a few weeks, it totally disappears from the peripheral blood, as regards ordinary routine examinations, while the animal shows no symptoms whatever.

When this trypanosome occurs together with *T. vivax*, or possibly with *T. gambiense*, death not uncommonly occurs after rapid emaciation and some paralysis have been observed. This, however, is not constant.

Tabulated details are given of the course of the disease in four goats, four calves, one bushbuck, and one sheep. Only one of the calves died, apparently from the infection. The death took place 18 months after inoculation (calf already mentioned).

The strain employed was originally obtained from a bushbuck, while that employed by BRUCE was obtained from oxen. This may possibly explain the difference of virulence.

Antelope as reservoir for T. uniforme.

In one out of a series of five experiments it was shown that a situtunga was capable of infecting *G. palpalis* with *T. uniforme* after a period of ten months, although the animal remained in excellent health.

(396) DUKE (H. L.). Further Investigations on the Rôle of Antelope as a Reservoir of *T. gambiense*. — *Reports of the Sleeping Sickness Commission of the Royal Society*. 1913. No. 13. pp. 58-66.

In a previous paper (see this *Bulletin* Vol. 1. No. 2. Abst. No. 135. pp. 94-95) experiments were described dealing with the duration of infection of antelope with *T. gambiense*, and the following conclusions were arrived at:—

1. The antelope may remain capable of infecting *G. palpalis* with *T. gambiense* for a period of at least 22 months after their original infection.

2. There is some evidence to show that an antelope which has ceased to be infective for *T. gambiense* acquires some degree of immunity against re-infection.

In the present paper the history of these antelope during the period March 1912 to November 1912 is followed out.

Reedbuck 2357. The blood of this animal had last been proved infective eight months previously. It was fed upon by a number of flies which were infected and of which eighteen had positive salivary glands. After the lapse of an interval to allow the trypanosomes to make their appearance in the blood this animal was tested with clean laboratory-bred flies and by inoculation of its blood into susceptible animals. The flies after feeding were transferred to a clean monkey which they failed to infect. One monkey out of five inoculated became infected.

Reedbuck 2431. Last evidence of infection obtained fourteen months previously. On two occasions this animal was fed upon

by infected flies (four flies with positive salivary glands). Clean flies fed upon it infected two monkeys upon which they were afterwards fed. One monkey out of a batch of five inoculated became infected.

In a small number of experiments with white rats the serum of this animal was found to have no protective action.

Reedbuck 2359. This animal which had shown no sign of infection for eight months was tested in a manner similar to that adopted in the previous cases and was found to be susceptible to re-infection.

Bushbuck 2371. No attempt was made to re-infect this animal but its infectivity was tested with negative results (Control).

Waterbuck 2378. The tests of infectivity and attempts to re-infect were carried out as before, and the serum of the animal was tested for protective properties. It was found that the buck had acquired a degree of immunity which enabled it to resist re-infection by positive *palpalis*. Its serum was found to have no protective powers (rat experiments).

Bushbuck 2328. This animal was subjected to two attempts to re-infect it with positive *palpalis* and in each instance without result. It further resisted inoculation with the blood of an infected monkey. Its serum was without protective properties.

- (397) CANADA: DEPARTMENT OF AGRICULTURE. Report of the Veterinary Director-General and Live Stock Commissioner, J. G. Rutherford, C.M.G., for the Year ending March 31, 1912. —480 pp. With 29 plates. 1912. Ottawa. [Sessional paper No. 15b—1913.]

Dourine.

Dourine still exists to a limited extent in Southern Alberta where its presence was first discovered in 1904. The figures show that the number of horses slaughtered during the period under review (18) is far below that slaughtered in any previous year, and attention is drawn to the fact that in five instances the infection was traced to a stallion imported direct from Iowa.

- (398) ROBERTSON (Muriel). Notes on the Life-history of *Trypanosoma gambiense*, with a Brief Reference to the Cycles of *Trypanosoma nanum* and *Trypanosoma pecorum* in *Glossina palpalis*.—Reports of the Sleeping Sickness Commission of the Royal Society. 1913. No. 13. pp. 119-142. With 6 plates.

An abstract of the portion of this paper dealing with *T. gambiense* has already appeared in this *Bulletin* (1913. Vol. 1. No. 3. pp. 154-155.); the short section dealing with *T. nanum* and *T. pecorum* in *G. palpalis* may therefore be considered.

The development of *T. nanum* in *G. palpalis* has many features which closely resemble the conditions observed in connection with *T. gambiense*. The infection starts in the hind gut and by the 10th day there is an extensive invasion of the hind and middle gut. The method of division corresponds almost exactly with that observed in *T. gambiense*. Slender forms appear at the

10th to 14th days, and the proventriculus is invaded by the 20th day. The trypanosomes present in the proventriculus are not so uniform as in *T. gambiense* and the changes observed in the nucleus of the latter trypanosome do not occur in *T. nanum*. By the 25th day the parasites have invaded the proboscis and there assume the crithidial form.

The gut forms do not attach themselves to the gut wall, and crithidial forms are never observed in the gut cycle. The salivary glands are never invaded by this trypanosome.

The cycle of development of *T. pecorum* in *G. palpalis* is very slow, and transmission difficult to effect. Duke's experiments indicate that the fly is at the most a facultative host of the trypanosome. The course of development resembles that of *T. gambiense* and *T. nanum*, but the flagellates are larger and more massive than in the two organisms just mentioned. The slender forms are extraordinarily long and the nuclear changes seen in *T. gambiense* occur in this parasite. The first invasion of the proventriculus was observed on the 45th day of the cycle, and no proboscis infection was found before the 76th day. The proboscis infections were generally slight. The salivary glands never became infected.

- (399) DUKE (H. L.). *Some Attempts to transmit Trypanosoma gambiense by Wild Stomoxys; with a Note on the Intestinal Fauna of these Flies.*—*Reports of the Sleeping Sickness Commission of the Royal Society.* 1913. No. 13. pp. 89-93. With 36 text-figures.

The wild flies used in these experiments were *S. nigra* and *calcitrans*. These flies were fed upon infected monkeys and then placed upon clean monkeys. In no instance did infection result. Although no flagellates having crithidial or trypanosome structure were found in any one of the flies dissected, a flagellate showing the structure of Bodo and a crescent shaped body were seen.

The latter appear to be non-motile and are often present in great numbers in the gut of the flies and the possibility is suggested that they may represent some stage in the life history of some sarcosporidian parasite.

- (400) DUKE (H. L.). *Notes on Trypanosoma gambiense and Glossina palpalis.*—*Reports of the Sleeping Sickness Commission of the Royal Society.* 1913. No. 13. pp. 13-21.

- (A) *Observations on the Physiology of T. gambiense in G. palpalis.*

These experiments were undertaken with the object of ascertaining whether the nature of the blood ingested by flies during the earlier days of experiment had any effect upon the development of *T. gambiense* in them.

A series of paired boxes was taken and fed for the same period upon a monkey whose blood contained *T. gambiense*. Throughout the experiment both series received parallel treatment.

The following is an example of a pair of these experiments each of which served as a control to the other.

Experiment 358.

| Date. | Day of Experiment. | Procedure. | Result. | Remarks. |
|----------------------|--------------------|----------------------|---------|--|
| 1911. Sept. 3 ... | 1 | Fed on monkey 199. | | |
| " 4 ... | 2 | Starved. | | |
| " 5-25 ... | 3-23 | Fed on bushbuck 123. | — | 6 positive flies dissected. Trypanosomes in gut only. |
| " 26 ... | 24 | Starved... .. | | 1 positive fly died. Flagellates in gut and thoracic gut only. |
| " 27-Oct. 7 | 25-35 | Fed on monkey 390. | + | 76 flies alive on 30th day. |

At the final dissection six more positive flies were found, four of which showed virulent salivary infection. The number of infected flies was 11·5 per cent. of the total.

Experiment 357 (control to 358).

| Date. | Day of Experiment. | Procedure. | Result. | Remarks. |
|------------------------|--------------------|--------------------|---------|-----------------------------|
| 1911. Sept. 2-3 ... | 1 | Fed on monkey 199. | | |
| " 4 ... | 2 | Starved. | | |
| " 5-25 ... | 3-23 | Fed on fowl 364 | | 1 positive gut only. |
| " 26 ... | 24 | Starved. | | |
| " 27-Oct. 6 | 25-34 | Fed on monkey 391. | + | 44 flies alive on 30th day. |

At the final dissection one positive fly was found with numerous flagellates in the salivary glands. The number of infected flies was 2·6 per cent. of the total.

A summary of the results obtained in the series of experiments is tabulated as follows:—

| Experiment Number. | Percentage of positive flies with goat or buck blood. | Percentage of positive flies with cock or monkey blood. |
|--------------------|---|---|
| 357 & 358 | 11·5 | 2·6 |
| 567 & 568 | 3·5 | 0 |
| 581 & 582 | 20·6 | 3·6 |
| 617 & 618 | 3·7 | 0 |
| 127 & 128 | 0 | 3·3 |

The experiments summarised above represent all the work done in this connection up to the time of writing and have not been especially selected.

The results obtained are in marked contrast to those recorded by KLEINE and FISCHER who, using a far greater number of flies, obtained approximately 10 per cent. of infected flies when monkeys were used throughout the experiment and 2.4 per cent. when ruminants were used.

The possibility of the flies having picked up flagellates other than *T. gambiense* is practically excluded by the following control experiments.

Bushbuck 123 which had been infected with *T. uniforme*, a trypanosome readily transmitted by *G. palpalis* and confined exclusively to the proboscis, was tested on three separate occasions with a number of flies of which 180 were dissected with negative results. On two occasions its blood was used for the inoculation of clean monkeys again with negative results.

One of the sheep used in the experiments had never been infected at the laboratory and during a period of three months its blood was examined on 57 occasions with negative results. The other sheep used had been infected with *T. uniforme*, but although its blood had been examined from time to time for nearly a year no evidence of infection had been obtained.

KLEINE and FISCHER conclude that monkeys and man are far more important than ruminants as a reservoir for sleeping sickness, but the whole of the work done at Mpumu would point to the opposite conclusion.

(B) *To study the effect of different kinds of blood on the longevity of laboratory-bred G. palpalis.*

A table is given showing the details of ten experiments in which batches of flies numbering from 17 to 75 were fed for periods of two to four days upon different species of animals and then starved, the number of deaths in each batch being recorded daily. It is pointed out that the weak point in the experiment lies in the fact that it was impossible to obtain a number of flies of exactly the same age, and also to ensure satisfactory feeding of all the flies.

(C) *To prove that G. palpalis infective for T. gambiense is capable of infecting monkeys on three consecutive days.*

A batch of flies which had been shown to be infective was fed upon three monkeys on three consecutive days. Each of the monkeys became infected, and on dissecting the batch of flies only one was found to be infected, numerous flagellates being present in the gut and salivary glands. This fly must have infected all three monkeys.

(D) In a single experiment it was shown that the feeding of a large number of negative *Glossina palpalis* on a monkey does not interfere with the subsequent infection of the monkey with positive flies.

- (401) ROUBAUD (E.). *Relations Bio-géographiques des Glossines et des Trypanosomes.* [The Bio-geographical Relationship between Glossina and Trypanosomes.] — *Bull. Soc. Path. Exot.* 1913. Jan. Vol. 6. No. 1. pp. 28-34.

Having frequently noticed the inconstancy and infrequency of salivary infection in flies, the author formed the opinion that local climatic conditions might influence Glossina in such a way as to render them unfavourable for the evolution of the cycle of trypanosome development, and suggested that strains of flies differing from each other with regard to the development of trypanosomes in their salivary apparatus might make their appearance. In 1910 an experiment was made with *T. cazalboni* which justified the conclusion arrived at. Later, in conjunction with BOUET, the observations were confirmed and the "partial disinfection of the Glossina under the physical conditions of captivity" was established.

Experiments with Glossina soon showed that if a batch of flies be given infective feeds only a proportion acquire salivary infection, and that if the number of feeds be increased the number of flies showing salivary infection is also increased, but that there are always some that do not show this infection. The author states that for the acquisition of this infection the flies may have to be placed in certain physiological conditions which he calls "receptivity." This applies only to the salivary infection and may be independent of infection of the alimentary canal. It may be temporary and disappear at the end of a certain time.

Under natural circumstances it has been shown that Glossina of different species frequenting the same district are not infected in the same proportions, nor in the same manner. In Lower Dahomey *T. cazalboni* predominates in *G. longipalpis* and *palpalis*, *T. dimorphon* in *G. longipalpis* and *tachinoides*, *T. peccaudi* in *G. longipalpis*. In Upper Dahomey, *T. peccaudi* predominates in *G. morsitans*, and in Casamance *T. dimorphon* in *G. morsitans*.

In view of the fact that wild domesticated animals exist in these areas under the same conditions, the author thinks that the differences observed are due to bio-geographical differences to which the flies are subjected. The author's own observations regarding the variation of receptivity exhibited by *G. palpalis* towards *T. cazalboni* support this view.

At Brazzaville in the Middle Congo 4.8 per cent. of *palpalis* became infected in his experiments (41 flies given one infective feed). At Agouagon in Middle Dahomey the average percentage was 40, but it varied from 10, when there was a single infective feed, to 70, where several infective feeds were given. Repeated infective feeds failed to infect a single fly out of 60 at Kolda in Upper Casamance.

Examination of naturally infected flies confirmed these differences. In Middle Dahomey one fly in 30 was found to have a proboscis infection, while in Upper Casamance 560 flies were examined and only one with proboscis infection found. Souma was of fairly frequent occurrence in the area, but it must have been transmitted by some other species of Glossina.

Comparison is made between the results obtained in the experiments regarding the transmission of *T. gambiense*, and those obtained by BRUCE and KLEINE, the latter authors finding as many as 5 per cent. of flies infected in this way.

In Dahomey where the disease does not exist not one fly in 1,200 was found to be receptive.

Mention is also made of KINGHORN and YORKE's results with *G. morsitans* and *T. rhodesiense*, in their experiments made in the Luangwa Valley and on the plateau.

The failures of KLEINE and FISCHER on the Victoria Lake, and the successes of TAUTE on Lake Tanganyika are also contrasted.

The conclusion drawn by the author is that the receptivity of a given species of *Glossina* for a certain virus is not uniform throughout the area of distribution of the fly, and that receptive, refractory, and indifferent strains occur in nature.

The author thinks that it is incorrect to state that climatic influences exercise an effect upon the development of the virus in the flies, at any rate directly. Bio-geographical factors produce modifications in the saliva which are favourable or otherwise to the development of the trypanosomes. This is shown by the fact that in a given area certain species are receptive and others are not. All other factors being the same, it must be the receptivity of the flies that varies.

If then the receptivity of the flies varies it follows that disease caused by trypanosomes transmitted by them will only be endemic in districts where receptive flies exist. These districts, however, are not constant owing to migrations of the flies. This possibly explains the occurrence of sporadic cases of trypanosomiasis in districts where flies are numerous, but where trypanosomiasis is not of common occurrence. In such areas cases of disease often tend to be of a mild type.

The author has frequently observed this in cattle and in horses in Gambia and Casamance. It is conceivable that geographical factors may exert an influence upon infected *Glossina* passing from an endemic zone to an indifferent one, in the direction of attenuating the virus, and even leading to absolute disinfection of the fly. Changes of this kind have been observed in the laboratory.

To sum up, the receptivity of the different species of flies in a district must be ascertained before any conclusion as to the endemicity of a disease can be arrived at, and the presence of a large number of flies must not be taken as evidence that such an area is dangerous either to man or animals. The introduction of a virus into a district in which flies are present is not of necessity dangerous, and the author believes that the chances of the extension of a virus outside areas of normal receptivity are very slender.

Any rational campaign against the flies must therefore be directed against the areas of receptivity, and measures of deforestation should be preceded by experimental investigation of the receptivity of the flies in the area, or at least by an examination of their natural transmitting power.

- (402) ROBERTSON (Muriel). *Notes on the Polymorphism of Trypanosoma gambiense in the Blood and its Relation to the Exogenous Cycle in Glossina palpalis.*—*Reports of the Sleeping Sickness Commission of the Royal Society.* 1913. No. 13. pp. 94-110. With 26 charts.

This paper deals with the fluctuation of the number of trypanosomes present in the blood of an infected animal, and with the question of polymorphism.

Attention is drawn to the evidence supporting the view that multiplication of the parasites occurs in the circulating blood and that it is improbable that invasion of the cells of the host for the purpose of multiplication occurs. The factors governing the number of trypanosomes present in the blood at any given time are discussed at some length.

The preparations used in the investigations of the polymorphism of the parasite were fixed first with osmic acid vapour and then with alcohol, being afterwards stained with Giemsa. The parasites were measured, after having been drawn with a camera under a magnification of 2,000, by BRUCE's method.

There is no sharp division of trypanosomes into types, the forms merging into each other. It is believed that the short forms represent the "adult blood type," so called because it is the most stable form. These become elongated and more slender and, passing through the intermediate stage, reach the long slender form. These long forms divide to produce the short forms again.

The question whether any special conditions of the parasite or of the infection are essential for the production of positive flies is considered and the view is held that an average number of positive flies is obtained when there has been a drop in the number of trypanosomes in the blood and those present are mostly of the short type.

- (403) OGAWA (M.). *Quelques Observations sur le Dimorphisme de Trypanosoma pecaudi.* [The Dimorphism of *T. pecaudi*.]—*Centralbl. f. Bakt.* 1. Abt., Orig. 1913. Mar. 15. Vol. 68. No. 3-4. pp. 332-334. With 3 text-figures.

During the course of an infection with *T. pecaudi*, the cause of Baleri, two forms of the parasite are observed, one long and slender and the other short and stumpy. In his original investigations Pecaud was inclined to think that he was dealing with a double infection, but was never able to furnish proof of this.

The author has made observations with the object of ascertaining whether any connection exists between the course of the disease and the occurrence of the two forms of parasite in the blood, and for this purpose has adopted the biometric method. Long forms and dividing forms make their appearance in the blood some days before the short stumpy forms.

In animals like the guinea-pig in which the course of the disease is (as a rule) sufficiently long, the short forms occur in large numbers during the middle period of infection, while during the last stage the long forms again predominate.

In animals like the mouse, in which the course of the disease is rapid, the long forms outnumber all other forms during the whole period of infection.

Measurements were taken of trypanosomes occurring in the blood of two guinea-pigs and a chart constructed upon the results obtained.

The first guinea-pig survived until the 16th day after subcutaneous inoculation with blood containing a large number of parasites, and the second, inoculated in a similar manner, died on the 7th day. The measurements were made on smears fixed while moist with osmic acid and stained with Giemsa. In the case of the first guinea pig 1,000 trypanosomes were measured, and in the case of the second 200.

The long forms measured from 24 to 34 microns in length and from 1 to 1.5 in width. The short stumpy forms without free flagellum varied from 12 to 20 microns in length and from 2.5 to 4 in width. Intermediate forms provided with a short free flagellum measured 21 to 23 microns in length and 1.5 to 2 in width.

From the chart given it may be gathered that in the first guinea-pig there were trypanosomes varying in length from less than 12 microns up to 33 microns, whereas in the second guinea-pig the shortest trypanosome encountered measured 18.5 microns in length and the longest 32 microns. In both animals the trypanosomes measuring about 25 microns were the most numerous, but in the first the percentage of these was only just over 6, while in the second the percentage was 11.

The author's examinations of smears containing *T. pecaudi* enable him to support the statements made by WENYON, LAVERAN, and NATAN-LARRIER that posterior nuclear forms of *T. pecaudi* occur. In rare instances the nucleus was actually posterior to the blepharoplast.

The author has been unable to find any indication of sexual forms.

(404) DUKE (H. L.). A Trypanosome from British East Africa Showing Posterior-Nuclear Forms. (With a Note on its Developmental Stages in *G. palpalis*. By Muriel ROBERTSON.)—*Reports of the Sleeping Sickness Commission of the Royal Society*. 1913. No. 13. pp. 67-89. With text-figures.

This organism, which was obtained from MONTGOMERY, was originally derived from a donkey which was supposed to have contracted the disease at a place a day's march on the road from Ngobotok to Baringo. According to P. Ross tsetses are known at three places on that road and the species found are *G. pallidipes*, *G. longipennis* and *G. brevipalpis*. The disease is described by CROWTHER as lasting four days.

In the living state the trypanosome executes very active and ceaseless wriggling movements with the anterior portion of the body while the posterior portion may remain almost motionless. There is very little translation, but when this does occur the movement is of a steady gliding nature.

In fixed and stained preparations a varying number of granules may be seen in the body. These may be anterior or posterior to the nucleus, or scattered throughout the body. They are most frequently observed anterior to the nucleus.

The kinetonucleus is as a rule not quite terminal, and on rare occasions it may be seen unusually far forward.

The trophonucleus is in a very considerable number of cases situated in the posterior portion of the body, and often quite close to the posterior end. This arrangement is more frequently observed in the short and intermediate forms. The proportion of posterior-nuclear forms varies in different species of hosts. They are relatively common in the dog and rat, and rare in monkeys. Occasionally they may number thirty per cent. of the parasites present.

The undulating membrane is well marked.

In the long forms there is a long free portion of the flagellum. The stumpy forms usually have a short free portion, but in some the protoplasm extends to the tip. In certain specimens a clear vacuole measuring as much as three microns in diameter is seen just anterior to the kinetonucleus.

On one occasion an individual was seen in the circulation of a dog which suggested a process of multiple division.

In a certain proportion of specimens a staining line may be seen passing from the kinetonucleus to the trophonucleus, and thence to the anterior end of the body. The anterior portion of this line may sometimes shew bead-like thickenings. In wet preparations stained by Heidenhain's method the line is also visible, but the process of differentiation may be carried too far to allow either the line or the granules to be seen.

When the trypanosome was first sent to Mpmu MONTGOMERY mentioned that it was supposed to be *T. pecaudi*. This is of interest in view of WENYON's statement regarding the occurrence of posterior nuclear forms in *T. pecaudi* (see this *Bulletin*, 1913, Vol. 1, No. 2, Abst. No. 140, p. 99).

The trypanosome varies from 12 to 38 microns in length, and is markedly dimorphic.

The value of the biometric method is discussed at some length and some comparisons instituted, the conclusion being drawn that at present the method is unsatisfactory for the diagnosis of species.

Tables are given shewing the results of infection with this trypanosome of a number of different species of animals, and it is seen that broadly speaking the reactions obtained agree with those obtained by LAVERAN, CAZALBOU, and BOUFFARD with *T. pecaudi*. Guinea-pigs, however, appear to react in an exceptional manner. Five were inoculated from an infected dog and in one only were trypanosomes found. The period of incubation was 44 days. BOUFFARD's observations furnish a parallel to this, for he found that with *T. pecaudi* the period of incubation was never less than 20 days and in one instance was 53 days. An attempt to infect guinea-pigs with positive *G. palpalis* failed.

Although the general reactions of the trypanosome most closely agree with those of *T. pecaudi* (BOUFFARD's strain) the possibility

is considered that the parasite may be either a mild strain of *T. brucei* or *T. rhodesiense*. The lack of objective symptoms in sheep and goats appears to exclude *T. rhodesiense*, as does the general scale of virulence.

Experiments made with human serum shewed that the trypanosome is susceptible to the action of this serum either as a curative or as a protective, but the former action would appear to be weak.

Though the protective action is well marked complete protection was not observed.

The trypanosome can be transmitted by *G. palpalis* and it would appear that in infective flies the trypanosomes occur in the gut and salivary glands, but not in the proboscis.

The question of the diagnosis of species of trypanosomes by a study of the distribution of the parasites in the intermediate host is considered at some length. The trypanosome described in this paper belongs to the gambiense-rhodesiense group in that it develops in the gut and the salivary glands and not in the proboscis of *G. palpalis*. The *T. pecaui*, to which it bears some resemblance in other respects, belongs to the group of trypanosomes which develop in the gut and the proboscis of the transmitting flies—"infection totale" of ROUBAUD.

Note on the Life Cycle of the Trypanosome in the Alimentary Tract of Glossina palpalis.

The development of this trypanosome in *G. palpalis* is in the main very like that of *T. gambiense*, but there are certain differences which enable the two to be distinguished.

The earliest evidence of development was seen in a fly on the third to fourth day after infection. The trypanosomes had increased somewhat in size and bulk and were extraordinarily uniform in type. Division was in active progress and the parasites had spread through the mid and hind gut. The details of division are identical with those of *T. gambiense* (see this *Bulletin* 1913, Vol. 1, No. 3, Abst. No. 285, pp. 154-155). It is stated that the details of division of the four trypanosomes *T. nanum*, *T. pecorum*, *T. gambiense*, and the organism at present under consideration correspond so closely that one set of figures would correctly illustrate the process in any one of them.

By the seventh day the trypanosomes in the mid and hind gut begin to lengthen. Certain very characteristic types are found about this period; they are distinguished by the body being drawn out rather sharply and stiffly posteriorly, and in the living state they have a peculiar stiff vibratile motion. About the fourteenth day long slender trypanosomes appear in the proventriculus where they form the predominant type.

At this stage there is often a period during which no further development takes place, but after this interval the slender proventriculus forms may be found in the hypopharynx and then in the salivary glands. These forms are apparently not capable of infecting and the development of typical salivary forms appears to be absolutely necessary.

The slender forms attach themselves in or near the duct. They then become broader in shape, divide, and migrate backwards along the gland where they re-attach themselves. They

assume the crithidial state and become quite rounded in shape. Multiplication goes on and this as a rule appears to be binary. The gland becomes crammed with parasites and the crithidial forms gradually develop into trypanosomes. This takes place about the 40th day. No evidence of conjugation has been observed in any phase of the cycle.

The features in which the cycle differs from that of *T. gambiense* are:—

1. The form under discussion is characterised by a rapid and numerous early development in the mid and hind gut, the early infections showing far greater numbers of parasites than *T. gambiense*.

The type of trypanosome present at this period shows a striking uniformity in shape and size.

2. There is in the trypanosome here described a marked pause between the first appearance of the trypanosomes in the proventriculus and their invasion of the salivary glands, which may amount to as much as 15 to 20 days.

3. The gland infections show far higher numbers than in the cycle of *T. gambiense*, but the development in the glands in the strain under observation occupied a considerably longer period than is the case with the sleeping sickness organism.

(405) BLACKLOCK (B.). A Study of the Posterior Nuclear Forms of *Trypanosoma rhodesiense* (Stephens and Fantham) in Rats.—*Ann. Trop. Med. & Parasit.* 1913. Mar. 31. Vol. 7. No. 1. pp. 101-112. With 1 text-figure.

The experiments detailed in this paper were carried out with the object of determining the time of appearance of posterior nuclear forms in the peripheral circulation of rats and the numerical relationship which these forms bear to other forms of trypanosomes present in the blood from day to day.

A series of four groups of rats were inoculated intraperitoneally with measured doses of trypanosomes and the average duration of the period of incubation and of the course of the disease noted in each case. It was found that no definite variation of these periods could be discovered in the different groups, in spite of the fact that the dose in one group was eight times as large as that given in another group (8 million and 1 million). The average period of incubation was 4.5 days, and the average duration of the disease 15.5 days. Daily examinations were made of the blood, and in each film 200 trypanosomes were counted. In counting these every type of parasite whether dividing or not was included, and the number of trypanosomes showing posterior nuclei was noted. The posterior nuclear forms were classified according to the position occupied by the nucleus, and three groups were made: A, those in which the nucleus was only just posterior to the middle point of the body; B, those in which the nucleus was placed midway between the mid-point of the body and the posterior end; C, those in which the nucleus was quite close to the blepharoplast.

The first trypanosomes which appeared in the peripheral blood were of the long and slender, or intermediate types. Subsequently

short forms made their appearance, and after a few days posterior nuclear forms. The A form appeared first, then the B form, and lastly the C form. Occasionally A and B forms appeared together, and also A and C forms made a simultaneous appearance, but in no case was the C form alone observed as the first type of posterior nuclear form to occur in the blood.

From the counts made it appeared that the posterior nucleated forms increased in numbers from the time of their appearance, both actually and relatively to the other forms of the trypanosomes.

The constancy of the results obtained by counting 200 parasites in each film was controlled by counts made of a thousand trypanosomes in each daily film from two of the rats in each group. The results obtained confirmed those obtained in the preliminary counts.

In several rats from which films were made after death it was observed that the proportion of posterior nuclear forms to other forms increased considerably.

The various views that have been expressed to account for posterior nuclear forms are considered at some length. These explanations are:

(a) The occurrence of such forms as a constant constituent of certain strains.

(b) A mixed infection.

(c) Certain unexplained influences in the blood environment affecting the parasites.

(d) The transmitting agent.

The arguments against the first of these explanations are weaker than those against the others.

The occurrence of posterior nuclear forms in animal trypanosomes is referred to, and it is said that "the fact that such forms are found in animal strains does not diminish their claim to attention, since animals are known to be infected with human trypanosomes. The argument, that if so many animal strains are capable of infecting human beings there would be a great amount of human trypanosomiasis in regions where it does not at present abound, is not conclusive. It has to be definitely proved that there is actually no human trypanosomiasis in such animal-infected regions. The experience in Nyasaland, and in Rhodesia, Northern and Southern, renders it necessary to be cautious in considering a region free from sleeping sickness."

(406) STEPHENS (J. W. W.) & FANTHAM (H. B.). Further Measurements of *Trypanosoma rhodesiense* and *T. gambiense*. — *Ann. Trop. Med. & Parasit.* 1913. Mar. 31. Vol. 7. No. 1. pp. 27-39.

This paper contains detailed measurements of 1,000 trypanosomes (non-dividing) taken from a single rat infected with *T. rhodesiense* and from another infected with *T. gambiense*, the measurements being taken, 100 daily, on consecutive days.

In each case the whole of the figures are given, and from them various tables are constructed.

In Table III. the trypanosomes are classified in Bruce's three groups (a) 13 to 21 microns; (b) 22 to 24 microns; (c) 25 microns and upwards, and it clearly shows that there is a great daily variation in the proportions of the different groups present in the blood, thus rendering a series of measurements made from preparations made from an animal taken at random liable to considerable inaccuracy.

The whole of the steps are repeated with *T. gambiense*; the measurements of 1,000 trypanosomes, measured 100 daily, from a single infected rat, and the tables of averages and groups as before.

From the table showing the group divisions of the trypanosomes it is seen that the daily variation of the percentage of the different forms present is even greater in the case of *T. gambiense* than in *T. rhodesiense*.

The measurements of both trypanosomes are represented graphically.

It is seen from the results obtained that the average, maximum, and minimum measurements of *T. gambiense* are a little greater than those of *T. rhodesiense* and that comparing the trypanosomes according to Bruce's groups the most marked differences lie in the percentage of short and long forms, the intermediate forms being present in about the same percentage in each case. Short forms predominate in *T. rhodesiense* to the extent of about 7 per cent. and long forms predominate in *T. gambiense* to about the same extent.

The authors state that there is a fair correspondence between their own curves and those constructed by Bruce, but none between their own and those constructed by Kinghorn and Yorke. They believe that these differences must be due to differences of method.

The authors have constructed three curves in all (1) based on 1,000 trypanosomes from various animals, but including 600 rat trypanosomes; (2) based on 600 trypanosomes from rats alone, where samples of 20 were taken on a number of days from different rats; (3) 1,000 trypanosomes taken 100 daily from one rat for ten days.

The three curves have this in common, that the peak of each is at 26 microns. Further, there is close agreement between curves (2) and (3).

The possibility is noted that trypanosomes taken direct from the natural vertebrate host or from the fly may have characters different from those that have been maintained in laboratory animals.

Conclusion:—

"We must admit that we had hoped to be able to distinguish between the two species, *T. rhodesiense* and *T. gambiense*, by measuring one thousand specimens of each organism. Though these biometric results are not sufficiently conclusive, we think that it is generally admitted that the two species are distinct.

Microscopically, the two trypanosomes are indistinguishable except by the posterior nuclear character of *T. rhodesiense*. We believe that a curve only expresses graphically what the eye can

appreciate under the microscope, and that if two trypanosomes cannot be distinguished microscopically, we shall not be able to do so by measuring them. However, provided that further experience enables observers to agree as to the best procedure, it is no doubt a great advantage to have a correct graphical expression for what is otherwise only an impression, although it may be a quite accurate one. Further, these measurements should not be regarded as useless, as they will undoubtedly form the basis (provided all the protocols are given) for a critical statistical investigation in the future."

In making the measurements the authors adopted their plan of projecting the trypanosomes directly on to a screen and tracing them, using the 'tangent line' method for the purpose of measurement.

- (407) FAVERO (F.). *Contribution à l'Étude de la Differentiation des Trypanosomes.* [The Differentiation of Trypanosomes.]—*Receuil de Méd Vét.* 1913. Jan. 15. Vol. 90. No. 1. pp. 10-13.

The author's investigations were carried out in order to test the conclusions arrived at by LEVADITI and MUTERMILCH.

According to these authors, leucocytes attack trypanosomes (alive or dead) only when the trypanosome used in the experiments in vitro is of the species which has been used for the infection of an animal, the serum of which has sensitised the leucocytes.

Favero's experiments had absolutely negative results in every instance, although the technique advised by Levaditi and Mutermilch was closely followed. Modifications of the technique did not lead to any better results.

LAVERAN and THIROUX, who have also tested the method, find that it has only a relative value, and that it is less accurate than the method of cross-immunity.

- (408) STEPHENS (J. W. W.) & BLACKLOCK (B.). *On the Non-identity of Trypanosoma brucei*, Plimmer and Bradford, 1899, with the Trypanosome of the Same Name from the Uganda Ox.—*Proc. Roy. Soc.* 1913. Mar. 6. Series B. Vol. 86. No. B 586. pp. 187-191.

In the introduction to this paper the authors briefly summarise the statements that have been published regarding the morphology of *T. brucei*.

BRUCE states that the trypanosomes vary a good deal in shape and size, and that they appear to vary a little in form in different species of animals. One or two of his figures may possibly represent stumpy forms.

The description given by KANTHACK, DURHAM, and BLANDFORD suggests dimorphism, but no mention is made of forms without free flagellum. No slides were available for examination, but from the examination of a large series of photographs (DURHAM) it was found that, save for one or two doubtful stumpy forms, the parasite was markedly uniform in shape and size.

PLIMMER and BRADFORD describe four forms, but neither their description nor figures suggest that stumpy forms were observed.

BRUCE and others state that after comparison of old Zululand preparations with preparations from the Uganda strain they consider them to be identical.

In another paper BRUCE states that *T. brucei* (Uganda strain) has 26 per cent. of non-flagellate forms.

LAVERAN classes *T. brucei* among the monomorphic trypanosomes.

The present authors have made a detailed examination of the two strains in a series of slides throughout the entire period of infection in various animals, viz., rats, guinea-pigs and rabbits.

The Zululand strain of trypanosome was derived from "a dog infected by the disease on the voyage from Africa, and brought to England in November, 1896, by Dr. Waghorn."

The Uganda strain was obtained originally from the Uganda ox in 1909. In 1912 this strain was lost for a short time, but was returned by MESNIL, who received it from Liverpool.

The authors state that they have established the following facts:—

(a) The Zululand strain is typically monomorphic, and the trypanosomes possess a long free flagellum. Short forms resembling true stumpy forms are to be found, but prolonged searching is necessary to discover them.

(b) LAVERAN's strain of *T. brucei* is monomorphic. Examination of old Zululand slides lent by NUTTALL, SKINNER, and PLIMMER show that the strain is monomorphic, although prolonged search reveals short forms, but these have not the somewhat indefinable characteristic appearance of true stumpy forms.

(c) The Uganda strain is typically dimorphic, true stumpy forms being easily found and occasionally in abundance.

The Uganda strain was kept by MESNIL in mice for nearly a year, and very few stumpy forms were to be found, but they immediately reappeared on the trypanosome being passed into guinea-pigs.

The authors suggest three explanations of the facts:—

1. That the strain that has been designated *T. brucei*, Zululand, is not this strain at all, but some other trypanosome inoculated erroneously during the course of inoculations extending over years. They do not think this view is tenable.

2. That BRUCE may have sent to England a strain other than the one with which he was working in Zululand. This is thought to be all the more likely as BRUCE was able to infect dogs from wild game, and there was no suspicion at the time of the multiplicity of animal trypanosomes in Africa.

3. That the strain originally sent to England was dimorphic, but that it has now become monomorphic. This may have come about in two ways:—

a. The strain was originally a mixture, the stumpy form having died out. They regard this view as not impossible, but cannot at present prove or disprove it.

b. The strain was originally dimorphic, but not a mixture, and that it has become monomorphic. Of such a change there

is not much evidence at present, but it has been noted by MESNIL, and confirmed by the authors, that the Uganda strain kept in mice for a year was almost, but not entirely monomorphic, but that in guinea-pigs it at once showed its normal characters.

The authors suggest that the Uganda trypanosome should be renamed *T. ugandae*.

- (409) LAVERAN (A.) & ROUDSKY (D.). Essais d'Immunisation contre les Trypanosomes pathogènes. — Trypanotoxines. [Attempts to Immunise against the Pathogenic Trypanosomes.—Trypanotoxins.] — *Bull. Soc. Path. Exot.* 1913. Mar. Vol. 6. No. 3. pp. 176-181.

This paper contains the results of some experiments in which the technique devised by BRAUN and TEICHMANN was followed save that the materials were prepared with aseptic precautions instead of sterilisation being effected by means of toluene. This was done because it was found that accidents occurred if every trace of toluene was not removed.

The material used for the immunisation was a powder composed of dry trypanosomes (*brucei*) which was suspended in salt solution. Each mouse received five doses of 2 cg. at intervals of five days, the inoculations being intraperitoneal.

In the first series of experiments an attempt was made to vaccinate eight mice, but each succumbed to a test inoculation with virus.

Two attempts to vaccinate two rabbits by a similar method also failed. The dose of dried trypanosomes given in these cases at each inoculation was 5 cg.

In the third batch of experiments serum was obtained from a rabbit which had received four intraperitoneal inoculations of 5 cg. of dried *T. brucei*. Two mice were given 1 cc. each of this serum by intraperitoneal injection, and on the following days were inoculated with blood containing *T. brucei* in very small numbers. Both of these mice died.

A third mouse was tested after the administration of the serum by inoculating it with *T. congolense*. This mouse became infected but apparently recovered. A control mouse inoculated with *T. congolense* became infected and was still infected at the time of writing.

In all the experiments control animals were inoculated with the test virus.

A second rabbit which received seven doses of powdered dry *T. brucei* yielded a serum which was not more active than the first, and the serum of this rabbit failed to protect when it was mixed with the virus at the time of the inoculation of the experimental mice.

The authors find that the method is of no value, and they point out that for their experiments on eight mice and four rabbits more than a hundred large rats had to be killed for the provision of the trypanosomes.

According to BRAUN and TEICHMANN *T. brucei* does not contain any toxin and here again the authors' observations are at

variance with those of BRAUN and TEICHMANN. Brief details are given of two experiments in which the intraperitoneal inoculation of two mice with 6 cc. of dried *T. brucei* caused marked symptoms of ill health with hypothermia in one and death with a lowering of the temperature of 5 degrees in the other. There was no peritoneal haemorrhage to account for the animal's death.

- (410) VON SCHUCKMANN (W.) & WERNICKE (K.). **Einiges über Methoden und Ergebnisse der Trypanosomenzüchtung.** [Methods and Results of Trypanosome Cultivation.]—*Centralbl. f. Bakt.* 1. Abt., Orig. 1913. Mar. 1. Vol. 68. No. 2. pp. 241-255. With 1 text-figure.

In this paper are described various modifications of the Novy-McNeal method of cultivating trypanosomes. Attempts were made to replace the rabbit blood by blood from other animals and also by using mixtures of serum from one species with corpuscles from another. The method was further modified in that the tubes of media were sterilised by heat after the processes of preparation were complete, in order to destroy any contamination that might have gained access during the defibrination of the blood.

A table is given in which the results of sixty experiments are set out. This table shows the nature of the medium, its age, the number of tubes inoculated, the results, the period for which the cultures remained alive, and the number of generations through which each was carried.

For the majority of the experiments bird trypanosomes were used.

It was found that no particular blood or serum was especially favourable for the cultivation of the parasites.

Details are also given in tabular form of a large number of culture experiments made to control the correctness of SCHAUDINN's view regarding *Haemoproteus* and *T. avium*. The authors were unable to obtain any evidence in support of his conclusions and they are unable to agree with the view expressed by MAYER who claimed to have proved the accuracy of SCHAUDINN's observations.

- (411) PONSELLE (A.). **Recherches sur la Culture *in vitro* du Trypanosome de l'Anguille (*Trypanosoma granulolum* Laveran et Mesnil 1902). Une Nouvelle Modification au Milieu de Novy et MacNeal.**—[The Cultivation *in vitro* of the Trypanosome of the Eel.]—*Compt. Rend. Soc. Biol.* 1913. Feb. 21. Vol. 74. No. 7. pp. 339-341.

Referring briefly to the various publications regarding the cultivation of this trypanosome the author states that he has tried the NNN medium and the heated blood medium of MATHIS, but has never obtained more than a prolonged preservation of the parasite as it occurs in the blood, without any indication of multiplication. In one instance the parasite was maintained for

more than a month. The nutritive value of the media tried has been determined in each case by making cultures of *T. lewisi*.

A modification of the NNN medium has however been found in which *T. granulosum* grows rapidly, and in which the forms observed in the intestine of *Hemiclepsis marginata*, the intermediate host, have been developed.

The composition of the medium is as follows:—

Unwashed agar 20 grammes.

Ordinary water 1,000 cc.

The agar is melted and filtered and distributed to tubes in quantities of 2 to 3 cc. and then sterilised. After cooling to 50° C. an equal volume of defibrinated rabbit blood is added and the medium allowed to set in the slanting position. The culture medium is the water of condensation.

The feature of the medium is the omission of sodium chloride, rendering it very hypotonic as compared with the blood of vertebrates. The presence of 4 per 1,000 sodium chloride does not absolutely inhibit growth, but very few of the trypanosomes divide.

(412) NATTAN-LARRIER (L.). Contribution a l'Étude de l'Action de la Bile sur les Trypanosomes. [The Action of Bile upon Trypanosomes.]—*Bull. Soc. Path. Exot.* 1913. Jan. Vol. 6. No. 1. pp. 24-28.

The experiments were carried out with the acentrosomic variety of *T. brucei* (Werbitzki), and mice were used in the majority of instances. The trypanosomes were obtained by centrifuging citrated blood, removing the trypanosome layer, and washing with salt solution. The bile was, in most cases, fresh ox bile which had been passed through a Chamberland filter. In the immunisation experiments, however, desiccated bile was used, the bile being dissolved in salt solution. The "virus" used was the sediment of washed trypanosomes mixed with an equal volume of salt solution.

In the first series of experiments the action of bile upon trypanosomes was tested.

A series of tubes was prepared by mixing two drops of "virus" with ten of salt solution and then adding bile to each tube, the first receiving two drops, the second three, and so on. A control tube was kept (the first tube in the author's series).

Preparations were made from each of the tubes after an interval of 15 minutes.

No motile trypanosomes could be seen by direct examination of a preparation from the first tube. In the other tubes, up to the seventh, the results were found to be "doubtful," while in the seventh, eighth and ninth no trypanosomes could be seen at all.

The contents of tube 1 proved fatal by intraperitoneal inoculation into a mouse. The contents of the ninth tube of the series failed to infect. The results of inoculation with material from the other tubes of the series are not given.

In the second experiment, blood containing a large number of trypanosomes was placed between a slide and cover-glass, a 60 per cent. solution of bile placed at one edge of the cover-glass, and the slide set aside at room temperature for 15 minutes. At this interval the trypanosomes were found to be still motile and apparently normal. After 20 minutes the movements were slowing down and after a further five minutes all movement had ceased, and the trypanosomes had a large posterior vacuole. After half an hour's exposure to the bile the outlines of the trypanosomes were ill-defined, and shortly after the parasites appeared granular and shrunken.

The third experiment reproduced the second save that the preparations were fixed and stained with Giemsa.

In the first preparation the majority of the trypanosomes were well stained, but some were swollen and stained of a faint pink colour. In the subsequent preparations various stages of degeneration were observed, until in the sixth the flagellum was practically all that remained, there being a few chromatin-like granules adhering to it, which no doubt represented the last remains of the cytoplasm and nucleus.

The second series of experiments was directed towards the treatment of trypanosomiasis by means of bile.

By experiment it was found that the toxic dose of bile for a mouse weighing 20 grammes was about 0.5 cc.

In this series of experiments the trypanosome of surra was used. A control mouse died on the eighth day. A second mouse was inoculated on the seventh day after infection, when trypanosomes were "not scanty" in its blood, with 0.5 cc. of bile. The mouse died two hours later, trypanosomes being present up to the time of death.

The third mouse received twice the dose of bile, and died in three hours, the trypanosomes again persisting up to the time of death.

In a second experiment a mouse in whose blood trypanosomes were scanty was given 0.5 cc. of bile. Trypanosomes were not again observed in its blood during the day, but the animal died on the following day.

In the third series of experiments an attempt was made to immunise mice by administering successive doses of a mixture of "virus" and bile, which had been incubated at 38° C. for 15 minutes, the quantity of bile in the successive doses decreasing. In no case was it possible to show that the mice acquired any immunity.

(413) HOLMES (J. D. E.). *Salvarsan in the Treatment of Surra in Horses.*—*Memoirs of the Dept. of Agric. in India, Veterinary Series.* 1913. Vol. 1. No. 2. pp. 89-107.

In preliminary experiments the toxic dose by intravenous injection was ascertained approximately, and the effects of both small and full sub-toxic doses were gauged by the period for which the peripheral circulation remained free from trypanosomes after treatment.

In the first series of experiments 17 animals were given a single dose of the drug. Of these three recovered, 11 had relapses, and three died. The dose administered varied from '0014 to '0094 grammes per pound body-weight.

In the second series of experiments three injections were given at intervals of one day. Only two animals were treated in this manner; in one there was a relapse and the other remained under observation for ten months without a reappearance of the trypanosomes being recorded.

Two animals received three injections at intervals of one week. In both cases there was a relapse. Four animals were given, in addition to the three injections of salvarsan, one dose of arsenic *in bolus*, the dose being 1 gramme. In none of these was a cure effected.

The author's conclusions may be summarised as follows:—

1. The toxic dose of salvarsan for the horse administered intravenously is approximately '01 gramme per pound body-weight, but individual susceptibility exists.

2. An intravenous injection of salvarsan is followed by a disappearance of trypanosomes from the peripheral circulation for periods varying from 6 to 36 days. The relationship between the dose of the drug and the period of absence of the trypanosomes is neither regular nor definite.

3. In three cases a single dose effected a cure.

4. By repeated administration a tolerance to the drug is established.

5. Three intravenous injections did not give results superior to those obtained by a single injection.

6. The administration of a large dose of arsenious oxide followed 24 hours later by an intravenous injection of salvarsan produced no better results than a single dose of either.

(414) HOLMES (J. D. E.). *Salvarsan in the Treatment of Surra in Dogs*.—*Memoirs of the Dept. of Agric. in India, Veterinary Series*. 1913. Vol. 1. No. 2. pp. 109-146.

The difficulties in the way of treating dogs for surra lie in the facts that the dog is particularly susceptible to the disease and also very intolerant of the drugs which have given the most encouraging results in other species.

Twenty-nine dogs were given a single injection of salvarsan in doses varying from '006 to '063 grammes per pound body-weight.

Of these two remained under observation for 370 days without relapse. Eleven died in periods varying from a day to several weeks after treatment, but for the most part trypanosomes were absent from the circulation during that time. In four cases there were relapses, and the remainder of the dogs had to be destroyed before the experiments were complete owing to their having come into contact with a dog suspected of rabies.

In all these cases the drug was administered in a 0·5 per cent. solution.

In six instances the drug was used in a 1 in 250 solution, but all the dogs died within 48 hours after the injections were given.

Six dogs received three injections at intervals of a day, but either death or relapse occurred in each case.

Of two dogs which received three injections at intervals of a week one recovered and one died. Similar results attended the experiments when only two injections were given at a similar interval.

Three dogs were given a single subcutaneous injection but none recovered. It was found that subcutaneous injection produced painful swellings which either burst or were absorbed.

Intramucular injection proved successful in one case and failed in another.

The following conclusions are drawn:—

1. The toxic dose of salvarsan for the dog is about '025 gramme per pound body-weight, and '02 gramme is fairly well tolerated.

More dilute solutions produce more markedly toxic symptoms than more concentrated ones.

2. An intravenous injection may cause the disappearance of the trypanosomes from the circulation for periods varying from 20 to 97 days.

3. Repeated injections do not give better results than a single administration of the drug.

4. Subcutaneous and intramuscular injections are not well borne. Three rabbits which were given doses of '000042 to '000064 gramme intravenously all recovered.

(415) SALMON (P.). *L'Acridine dans le Traitement de la Maladie du Sommeil expérimentale.* [Acridine in the Treatment of experimental Sleeping Sickness.]—*Compt. Rend. Soc. Biol.* 1913. Jan. 24. Vol. 74. No. 3. pp. 134-136.

A number of observers have recorded the effects produced by certain dyes of the acridine or oxazine series upon trypanosomes. There is vital staining of the blepharoplast, followed by disappearance of this structure, and destruction of the parasite.

During the course of a number of experiments regarding the process by which these changes are produced the author has obtained some favourable results in the treatment of trypanosomiasis, and especially with *T. gambiense*.

The strain of *T. gambiense* used was one which constantly proved fatal to mice in six days. The dye which gave the best results was Trypoflavin A (acridine) administered subcutaneously.

A dose of 0.06 mg. was found to protect a mouse when injected simultaneously with infective blood. In one experiment, in which the curative action of the dye was tested, four mice showing numbers of trypanosomes in their blood were injected with doses varying from 0.25 to 0.04 mg. and all save the one which received the smallest dose recovered. The latter still showed trypanosomes in its blood on the seventh day of the disease. The control mouse died on the sixth day.

Experiments showed that there is a large margin between the toxic and the therapeutic dose, the proportion being about 5 to 1.

If, however, the toxicity of this dye be compared with that of the various anilin dyes, trypanflavin falls into the class comprising the dyes of fairly high toxicity; care should therefore be taken in its application.

The destruction of the parasites occurs slowly. Trypanosomes may be found in the blood several hours after the injection. This is somewhat different to what happens in the case of arsenical compounds, which destroy trypanosomes within an hour.

A point in favour of acridine as compared with such substances as trypanblue is that it does not cause general colouration of the tissues; it produces, however, a yellow fluorescence in the urine.

(416) COLES (A. C.). Trypanosomes found in a Cow in England.—*Parasitology*. 1913. Jan. Vol. 5. No. 4. pp. 247-252. With 1 plate.

After summarising the literature that has appeared on the occurrence of trypanosomes of the *theileri* type in the blood of cattle, the author describes two specimens found by him in the blood of a cow in Dorsetshire.

At the time when the trypanosomes were found the animal was the subject of redwater, *Babesia bovis* being present in the blood and about 2 per cent. of the blood corpuscles invaded. Two parasites were found in one blood film out of seven examined, and none were found in 20 films made two days later.

The trypanosome measured 98 microns in length including the flagellum. The body measured 88; the flagellum 10; from the posterior extremity to the kinetonucleus 37, and to the centre of the trophonucleus 44; the diameter of the body 6; and the width of the undulating membrane 3.5 microns.

(417) CAZALBOU (M. L.). Observation d'un nouveau Trypanosome chez le Lapin. [A new Trypanosome of the Rabbit].—*Recueil de Méd. Vét.* 1913. Mar. 15. Vol. 90. No. 5. pp. 155-158.

The following details are given of an outbreak of disease occurring among some young rabbits at Rennes.

The animals when about three weeks old suddenly ceased to grow, rapidly lost condition, became affected with paralysis of the hind limbs, and died after a few weeks. The appetite remained good throughout.

Examination of the blood revealed the presence of a trypanosome, a single specimen being found in 15 blood smears.

The parasite was a very large one measuring about 80 microns in length, and having a free flagellum measuring 10 to 12 microns. The body at its widest part measured 8 microns, and an undulating membrane was distinctly visible opposite this wide portion of the body only. The nucleus was central and oval in shape. It appeared to be composed of homogeneous protoplasm.

The author failed to infect guinea-pigs by inoculation, and circumstances prevented him from inoculating rabbits, so that he was unable to establish the pathogenicity of the trypanosome. A number of references are given to records of the occurrence of trypanosomes in the rabbit.

SPIROCHAETOSIS.

- (418) DANULESCO (V.). *Essais de Culture du Spirille de la Poule.* [The Cultivation of the Spirochaete of the Fowl.]—*Compt. Rend. Soc. Biol.* 1913. Feb. 28. Vol. 74. No. 8. pp. 369-371.

The author has successfully applied the medium described by NOGUCHI to the cultivation of the Spirochaete of the fowl.

The infective blood is mixed with an equal volume of 1.5 per cent. citrate solution and distributed in quantities of about 10 drops to tubes in which have been placed fragments of fresh rabbit kidney. Ten or fifteen cubic centimetres of unfiltered sterile ascitic fluid are then added and finally a layer of sterile vaseline. The tubes are incubated at 37° C. Absolute asepsis is essential.

Four samples of ascitic fluid were used in the experiments and one was found to be far superior to the other three.

The author has found that in citrated blood maintained at 37° C. the spirochaetes will remain alive for 24 to 36 hours, for 48 hours in broth containing a fragment of fresh tissue, slightly longer in simple ascitic fluid or ascitic agar, and for fifteen to twenty days in the medium described.

In the latter medium the multiplication of the parasites is rapid. Within twenty-four hours there may be 15-20 spirochaetes in each field of the microscope where immediately after inoculation only four or five could be seen. During the following three days the number may be more than doubled. The spirochaetes preserve their motility and their general appearance save that they are as a rule a little shorter than those occurring in the blood of an infected bird. During the first day the parasites are present in all parts of the liquid and especially around the fragment of kidney. Gradually they begin to collect into little masses, and as the cultures get older show a tendency to pass into the upper layers of the liquid.

From the fifth day, and sometimes even earlier, some of the spirochaetes present a deformed and irregular appearance, and they may also appear to be granular. The granular parasites have lost their motility. In the majority of the tubes the parasites are all dead by the twelfth day, but sometimes they survive until the 15th day and in one case a culture was still alive on the 20th day.

Subcultures have been made using as seed material cultures from four to seven days old. The author has succeeded in obtaining five generations of subcultures. In the later generations an increasing number of the tubes proved sterile and there was a steady decrease in the abundance of the growth.

- (419) LAUNOX (L.) & LEVADITI (C.). *Nouvelles Recherches sur la Thérapeutique Mercurielle des Spirilloses (Sp. des Poules et Syphilis du Lapin).* [New Investigations regarding the Mercurial Treatment of the Spirochaetoses.]—*Compt. Rend. Soc. Biol.* 1913. Jan. 10. Vol. 74. No. 1. pp. 18-21.

The group of compounds investigated by the authors in these experiments were the dinitro and diamino compounds of the mercury salt of dioxydiphenyl.

The toxicity and the instability of the mercury salt of dioxy-dinitrodiphenyl and of dioxydiaminodiphenyl rendered them unsuitable, and the diacetyl derivative of the second of these substances was the only one found to be of any value.

For the sake of convenience this substance is called No. 114. It contains 42.8 per cent. of mercury and the solution used was prepared by mixing 0.2 g. of substance 114 with 2 cc. of normal sodium hydrate and 18 cc. of normal salt solution.

For the rabbit the toxic dose by intravenous injection was found to be 0.04 to 0.05 per 1,000. The toxic dose was that which produced a fatal result in 15 to 20 days. The toxic dose for the fowl was practically the same.

It was found that the drug exercised a protective effect when administered intravenously in doses of 0.04 per 1,000 within 24 hours after infection. The curative effect is less marked, and the drug appeared to be more toxic for birds treated when parasites were numerous in the blood, owing to decreased resistance caused by the infection.

(420) GLEITSMANN. Beitrag zur Entwicklungsgeschichte der Spirochäten (Borrelia). [The Life-history of Spirochaetes (Borrelia).]—*Centralbl. f. Bakt.* 1. Abt., Orig. 1913. Feb. 12. Vol. 68. No. 1. pp. 31-49. With 1 plate and 2 text-figures.

After giving a somewhat lengthy review of the literature, the author describes his own investigations regarding the life history of the organisms, special attention being paid to the so-called "intra-corpuseular bodies" (BALFOUR).

Two strains were used, viz., MARCHOUX's Brazilian strain and the Sudanese spirochaete of BALFOUR.

The first series of experiments was carried out *in vitro* and the immunity reactions of the two spirochaetes with the homologous and the heterologous immune sera investigated.

The sera were added to the spirochaetes in dilutions from $\frac{1}{2}$ to $\frac{1}{200}$, and it was found in each case that the homologous serum exerted a far more powerful action on the parasites than the heterologous. A polyvalent serum tested with each of the spirochaetes exerted the same effect upon both.

Experiments carried out upon fowls indicated that in spite of the results obtained *in vitro* the parasites must be closely related since a fowl immune to one strain was also immune to the other. In these experiments fowls were inoculated with mixtures composed of blood containing the parasites and the homologous and heterologous immune serum. The birds remained healthy.

In a further series of experiments fowls were infected by means of ticks and the survivors reinfected with the other strain. In each case immunity had been established by the first infection.

The author then passes on to a consideration of BALFOUR's "intra-corpuseular bodies," specimens being examined with dark ground illumination and also in films stained in the manner recommended by BALFOUR. He rather favours the view that these bodies do not in reality represent a stage in the life cycle of the

spirochaete, but that they are degeneration products of some kind; although the possibility is admitted that they may represent some separate infection.

Tabulated details are given of the course of infection in fourteen birds. Three birds infected with the Sudan strain and one with the Brazilian strain showed chronic symptoms, but in no instance was the relapse associated with the presence of spirochaetes or intra-corpuseular bodies.

When the parasites were numerous present in the circulation the nuclei of the red corpuscles underwent a process of karyolysis and fragments which stained deeply were separated off. Spirochaetes were also observed to penetrate into red corpuscles without becoming altered in appearance.

LEISHMANIASIS.

(421) Row (R.). Some Experimental Facts *re* Kala-azar (Indian).
Second Memoir.—*Jl. Trop. Med. & Hyg.* 1913. Jan. 1.
Vol. 16. No. 1. pp. 1-2.

In a previous communication the author has published the results obtained by inoculating *Macacus sinicus* (see this *Bulletin*, 1913, Vol. 1, No. 3, Abst. No. 309, p. 173), and in the present paper records some further observations on this and other species of animals.

The monkey originally infected developed two lesions about the size of beans at the seats of inoculation after 5½ months. One of these was excised a fortnight after its appearance, and an emulsion made from a portion of it was used for the inoculation of fresh animals. Microscopic examination of the emulsion showed that it was not rich and that therefore the doses administered were very small. Two mice and a *Macacus sinicus* were inoculated intraperitoneally with the emulsion, and one mouse with a mere trace of juice obtained by puncture of one of the lesions of the first infected monkey. Two of the mice were killed six weeks and two months after inoculation and the third died in six weeks. In each case microscopic examination showed that there had been a generalised infection. The liver of the monkey was aspirated four and a half months after infection and the parasite was found in the blood in small numbers, both free and in macrophages.

The liver and spleen of the mouse that died were removed 8 to 10 hours after death and an emulsion made from them was injected intraperitoneally into a fresh mouse. When this mouse was killed six weeks later no evidence of infection could be found.

The livers and spleens of the other two mice were treated in the same way and in each case two fresh mice were inoculated. In none of these animals was any definite evidence of infection found, and the author thinks that these negative results may possibly be attributed to one of the following causes:—

1. Possibly the animals were examined too soon, although fully seven to nine weeks were allowed in each case.

2. The possibility of the attenuation of the virus by passage through mice.

3. The possible curative effect of the extract of the liver and spleen introduced simultaneously with the parasites.

In addition to the above results the author records the production of a local pinhead nodule in a *Macacus sinicus* inoculated cutaneously from the monkey mentioned in his first paper already referred to. The incubation period was five months. The nodule contained typical parasites which yielded flagellates in cultures.

Dogs, rabbits, guinea-pigs, and wild mice have been found refractory to small doses of the virus given intraperitoneally and to large doses of culture given intraperitoneally or subcutaneously.

- (422) GRAY (A. C. H.). *Leishmaniose naturelle du Chien à Tunis.* [Natural Leishmaniasis of the Dog in Tunis.]—*Bull. Soc. Path. Exot.* 1913. Mar. Vol. 6. No. 3. pp. 165-166.

Examinations made by other authors have shown that the proportion of infected dogs in Tunis is 1.6 per cent. The present author has examined 127 dogs and has found two infected (1.6 per cent.).

In one animal the parasites were numerous present in the bone marrow but scanty in the spleen and liver. Cultures were obtained on NNN medium, but inoculation into two dogs failed.

In the second dog parasites were very numerous in the bone marrow, spleen and liver. They were also found in the portal blood. Cultivation failed owing to the presence of impurities.

- (423) LIGNOS (A.). *L'Infection par Leishmania des Chiens de l'Île d'Hydra.* [Canine Leishmaniasis in the Island of Hydra.]—*Bull. Soc. Path. Exot.* 1913. Feb. Vol. 6. No. 2. p. 117.

It would appear from the statistics that the Island of Hydra is more extensively infected with kala azar than any other country in which the disease occurs. The total population of the Island is about 6,000 and the annual number of births about 120. It is found that one child in ten becomes affected.

In view of the facts, established by NICOLLE, that the disease occurs in the dog and that children can contract infection from that animal, an examination of the dogs in the Island was undertaken to ascertain in what proportion these were affected.

During the summer of 1912, 48 dogs were examined, this accounting for half the dogs on the Island. As the inhabitants objected to the destruction of their animals the examinations had then to be brought to a close.

Eight (16.66 per cent.) of the dogs were found to be infected.

The distribution of the cases during the months May to October was as follows:—

| | Dogs examined. | | | Infected. |
|---------------|----------------|-----|----|-----------|
| May ... | ... | ... | 5 | 1 |
| June ... | ... | ... | 16 | 2 |
| July ... | ... | ... | 8 | 1 |
| August ... | ... | ... | 10 | 1 |
| September ... | ... | ... | 7 | 2 |
| October ... | ... | ... | 2 | 1 |

- (424) LAVERAN (A.). Infections du Cobaye, du Lapin et du Chat par la *Leishmania infantum*. [The Transmission of *Leishmania infantum* to the Guinea-pig, Rabbit, and Cat.] — *Bull. Soc. Path. Exot.* 1913. Feb. Vol. 6. No. 2. pp. 110-114.

This article is a review of the various attempts that have been made to infect guinea-pigs, rabbits, and cats experimentally with *L. infantum*.

Guinea-pigs.—In 1909 the author in conjunction with PETIT shewed that the presence of the parasite in the peritoneum of guinea-pigs 59 days after intraperitoneal inoculation could be proved by cultural methods, flagellates being developed on NNN medium.

In 1911 FRANCHINI succeeded in producing a general infection in a young guinea-pig by intraperitoneal inoculation with 1 cc. of a fifteen-days-old culture of the eighth generation. The guinea-pig was killed on the 26th day. Parasites were found in smears from the spleen, liver, bone marrow, and blood, but more numerous in the spleen than elsewhere.

NICOLLE and BLAIZOT have failed to produce infection in spite of repeated attempts.

The author failed in the case of two guinea-pigs inoculated in the liver with material obtained from an infected dog, in a guinea-pig inoculated intraperitoneally with peritoneal serous fluid from an infected guinea-pig, and in four guinea-pigs inoculated in various ways with large doses of culture. In a single instance there was a slight localised infection of the peritoneum in a guinea-pig inoculated intraperitoneally with material from an infected dog.

Rabbit.—In 1911 VOLTINO published an account of a successful infection of a rabbit. The infection was achieved by placing some infective bone marrow from a dog upon the scarified cornea. Three months later there was opacity of the cornea and typical parasites could be found in large mononuclear leucocytes.

MANTOVINI produced a general infection by intravenous inoculation with a culture. The animal was killed 20 days after inoculation, and cultures were obtained. The progress of the infection in this case was very rapid.

NICOLLE, BLAIZOT and Laveran have failed to set up injection on a number of occasions.

Cat.—Apparently the only recorded case of natural infection in a cat is that described by Ed. and Et. SERGENT, LOMBARD, and QUILICHINI. In this instance a child, a dog, and a kitten aged four months in the same house were found to be infected.

NICOLLE examined 51 cats from Tunis and Gafsa without finding a single one infected. The same author inoculated four cats unsuccessfully. Laveran has made several attempts to infect cats but without success. NICOLLE and BLAIZOT failed to infect a cat by intravenous inoculation with growth from six tubes of culture.

While examining preparations made from fleas caught on cats inoculated but not infected with *Leishmania*, Laveran found large numbers of parasites bearing a close resemblance to *Leishmania*.

The organisms were round or oval in shape and measured from 2 to 6 microns in length, by 2 to 4 in width. In many cases they were grouped into clusters. The cytoplasm, which stained a faint blue with Giemsa, contained a round or oval principal karyosome and an accessory rod-like karyosome. Flagellated forms were not seen, but the author thinks that there can be no doubt that the parasite is related to the trypanosomides described by a number of authors as occurring in other species of flea.

- (425) BANDI (I.). Preliminary Note on the Identity of certain Leishmaniasis based upon Biological Reactions.—*Jl. Trop. Med. & Hyg.* 1913. Feb. 15. Vol. 16. No. 4. p. 50.

The primary object of the author's investigations was to ascertain whether a method of diagnosis based upon biological reactions could be devised.

In the first series of experiments an attempt was made to ascertain whether and in what measure specific substances are elaborated in the blood of animals inoculated with cultures of *Leishmania*. Two strains were used, one of *L. infantum*, and one of canine origin, both of which were obtained from NICOLLE.

Rabbits were used for the inoculation experiments.

The author has observed that repeated intravenous inoculations with cultures, while not giving rise to any pathological condition, induce the formation in the blood of specific substances which agglomerate the parasites, and which can be compared to bacterial agglutinins. The serum of inoculated rabbits shewed this property in dilutions up to 1 in 200. The serum of normal rabbits had no action.

The serum of rabbits inoculated with canine leishmania agglutinated both this parasite and *L. infantum* in dilutions up to 1 in 160. Sera of rabbits inoculated either with canine Leishmania or *L. infantum* agglutinated *L. tropica* in dilutions up to 1 in 70 only.

The fact that the serum of rabbits inoculated with *L. infantum* and with the canine strain agglutinate both the homologous and the heterologous parasite to the same extent while they agglutinate *L. tropica* to a much lower degree shews that there is a general group reaction, and also tends to support the view that the parasites of the disease in the dog and child are identical.

PROTOZOA.

- (426) NICOLLE (C.) & CONOR (M.). La Toxoplasmose du Gondi. *Maladie Naturelle.—Maladie Expérimentale.* [Toxoplasmosis of the Gondi—Natural and Experimental.]—*Bull. Soc. Path. Exot.* 1913. Mar. Vol. 6. No. 3. pp. 160-165.

In May 1912 the authors received a batch of Gondi from Matmata in Southern Tunis where the parasite was originally found in 1907 and again in 1908.

At the time of their arrival the animals had on them two species of ectoparasites,—an undetermined Ixode and a larval *Trombidium* of undetermined species. They were immediately freed

from these parasites. During the two months following their arrival half the animals died, and although a post-mortem was made in every case, *Toxoplasma* was not found. The mortality ceased in July, but the animals commenced to die again at the end of September, and by the beginning of January nine more were dead. Eight of these were found to be infected with *Toxoplasma*. Subsequently two more died but these were not affected.

On post-mortem the animals were generally found to be in fat condition, but in the majority of cases there were obvious lesions: a clear bilateral pleural exudate associated with congestion or hepatisation of the lungs, enlargement of the spleen up to three or four times, and occasionally slight enlargement of the liver.

The parasite was present in enormous numbers in smears from the spleen, less abundant in the liver and lungs, rare in the kidneys, and exceptional in the blood and bone marrow.

The parasites varied in shape and measured on an average 5 by 2.5 microns.

The nucleus was single and generally centrally placed. In those parasites in which it was placed towards one end that end appeared swollen, the other end being in many instances drawn out.

The parasites occurred either free or enclosed in cells. In the latter case mononuclear leucocytes were more frequently invaded than polynuclear leucocytes. Multiplication forms were very numerous, and the only form of multiplication observed was longitudinal division, the process starting at the nucleus. Whether intracellular or free, groups of parasites numbering as many as 20 may be seen and such groups generally have a rounded outline suggesting encystment. The authors do not agree with SPLENDORE that there is actual encystment for the following reasons:—They have never observed the stage preceding that in which every toxoplasma is individually recognisable, they have been unable to make out any envelope, and, save for differences in size, masses of parasites resemble each other exactly.

The authors have succeeded in infecting two gondis experimentally by intraperitoneal inoculation with bone marrow from an infected animal, the material used being very poor in parasites. The periods of incubation were eight and nine days.

The mouse is said by the authors to be more susceptible than the gondi, death taking place in five or six days after intraperitoneal inoculation. In the mouse, however, the infection is not a general one, the parasites being practically limited to the peritoneal exudate, of which there is a large amount, and to the endothelium of the peritoneal membrane.

In two cases the infection was successfully transmitted by intramuscular inoculation, the period of incubation being about a week.

Nicolle and MANCEAUX have already recorded one instance in which a guinea-pig was successfully infected experimentally. The present authors have had positive results in four out of seventeen animals.

Infection has only followed intraperitoneal inoculation and this fails unless young guinea-pigs are used. The period of incubation was four to six days. At the post-mortem there was found peritonitis with abundant exudate serous and fibrinous containing numerous parasites. There was an occasional organism in the liver and spleen, but none were found in the bone marrow and the blood.

The pigeon is susceptible to infection, but less so than the mouse, and the course of the disease is different.

Infection in the pigeon is slower and has a greater tendency to generalisation. Intraperitoneal inoculation is the only method of setting up infection. Of eighteen birds inoculated eleven died in the nine to thirty-one days, parasites being demonstrated in each case.

Parasites are present in the liver, in which they may be found in enormous numbers, spleen, lungs, bone marrow, kidneys and blood.

Toxoplasma was not present in the peritoneal false membranes save in those birds which died within a short period.

The authors have failed to infect rabbits, rats, a dog, monkeys, toads and frogs, and they have been unable to cultivate the parasite outside the body.

(427) LAVERAN (A.) & NATTAN-LARRIER. *Au Sujet des Altérations Anatomiques produites par le Toxoplasma cuniculi.* [The Lesions produced by *Toxoplasma cuniculi.*]—*Bull. Soc. Path. Exot.* 1913. Mar. Vol. 6. No. 3. pp. 158-160.

The authors have studied the macroscopic and microscopic appearances of the lesions in the liver, spleen, and lungs, in pieces of tissue received from SPLENDORE.

The spleen is markedly enlarged and the liver presents an appearance suggestive of acute miliary tuberculosis. The lungs have a number of greyish nodules scattered through them.

The lesions in the liver vary in size up to one millimetre in diameter, and are scattered throughout the organ. In the early stages the lesions are rounded, but later have rather ill-defined margins. Under the microscope it is seen that in the earliest stages there is a loss of detail and staining affinities on the part of the affected liver cells, the cells becoming necrotic. There is no influx of leucocytes into the lesion. In the neighbouring capillaries there is thrombosis and proliferation of the endothelial cells. The centre of the lesion becomes converted into a kind of fibrinous network in the meshes of which are to be seen nuclear fragments and large numbers of red corpuscles. Around this central portion there is an irregular zone in which the outlines of the cells are still distinct, but in which the endothelial cells of the capillaries are profoundly altered.

By means of ordinary staining methods the distribution of the parasite can be easily followed, the liver cells being to a large extent invaded. Eight to ten parasites may be frequently found in cells that appear to be quite normal, but others containing 20 to 30 parasites are in the early stages of necrosis. The neigh-

bouring capillaries may be normal or impermeable. In the latter case they contain immense numbers of parasites, but the endothelial cells are not invaded. The organisms vary from 20 to 25 microns in diameter, and are rounded or oval in shape. They contain a variable number of karyosomes. Toxoplasma is not to be found in the central portions of the lesions.

The lesions in the spleen and lungs have the same essential features as those in the liver.

- (428) RODHAIN (J.), PONS (C.), VANDENBRANDEN (F.), & BEQUAERT (J.). Note sur des Trypanosomides intestinaux d'*Haematopota* au Congo belge. [Intestinal Trypanosomides of *Haematopota* in the Belgian Congo.]—*Bull. Soc. Path. Exot.* 1913. Mar. Vol. 6. No. 3. pp. 182-184. With 1 text-figure.

The authors have encountered trypanosomides in the intestinal canal of two species of *Haematopota*.

Two flies out of a batch of 11 *Haematopota duttoni* (Newstead) were found to have flagellates and cysts in their rectum. The motile organisms resembled those described by BRUCE as parasites of *Tabanus secedens*. The cysts which were oval or pyriform presented an appearance similar to the spermoides described by CHATTON.

A quite different flagellate was found in *Haematopota vandenbrandeni* at Sankisia. In this instance only one fly out of 39 was found to harbour the parasite. Cysts were also found in this case.

The flagellates took the form of slender elongated crithidia, the posterior extremity of which was drawn out into a point, the anterior portion carrying an undulating membrane closely applied to the body.

The largest parasites measured from 45 to 50 microns in length, a free flagellum of 16 to 18 microns being included in this measurement. The body was between 1.5 and 2 microns in thickness at the level of the nucleus, which was situated at a point about 10 to 14 microns from the anterior end of the body. There was no distinct karyosome, but the nucleus contained a number of irregular granules of chromatin.

The blepharoplast was punctiform and was placed about four microns in front of the nucleus, and the flagellum was relatively thick.

Posterior to the nucleus there were often observed in the cytoplasm a number of metachromatic granules.

In addition to the large flagellates there were smaller forms measuring only 31 to 39 microns in length.

Between the crithidia provided with undulating membrane and flagellum and the cysts, there was a series of gregarine-like bodies with short flagella.

The cyst forms were about six microns by two to four microns and they were surrounded by a delicate eosinophile layer. One extremity was slightly larger than the other and contained the nucleus, just posterior to which was the blepharoplast.

Non-flagellated forms were also observed which appeared to represent the stage preceding cyst formation.

The name *Crithidia tenuis* is suggested for the flagellate found in *H. vandenbrandeni* of which it is probably a parasite proper.

(429) CHATTON (E.) & ROUBAUD (E.). Sporogonie d'une Hémogregarine chez une Tsétsé (*Glossina palpalis* R. Resv.). [Sporogony in a Haemogregarine in *G. palpalis*.]—*Bull. Soc. Path. Exot.* 1913. Mar. Vol. 6. No. 3. pp. 226-233. With 2 plates.

At Kolda on the Casamance the authors have encountered in the body cavity of *G. palpalis* the cysts of an unidentified haemogregarine. These forms have not been observed elsewhere nor in any other fly. Out of a total of 465 flies only four were found to be infected, and it should be noted that the intestinal tract of these four flies also contained *T. grayi*. The flies when fasting were distinguishable from others by the fact that the abdomen was slightly swollen, and on making an incision there was found a mass of whitish powdery material composed of rounded grains of different sizes. The largest had a mulberry-like appearance and contained a number of secondary cysts which in turn contained numerous sporozoites. Examined in salt solution and after slight pressure had been exerted vermicular bodies capable of exercising slow movements were found to escape. These bodies exactly resembled haemogregarines as they occur in the blood. The larger cysts were situated immediately under the chitinous covering of the body, and in the deeper parts among the loops of the digestive canal mature sporozoites liberated from the large cysts were to be found. Parasites were not found in the thorax, but both cysts and sporozoites were found in the head. The wall of the intestine and the salivary glands were free from parasites.

The authors have been able to follow the development of the sporozoites from the sporoblasts, but as yet they have not traced any copulation.

The youngest stage that they have observed is a rounded body measuring from 50 to 70 microns. This is surrounded by a thin cyst membrane and contains a large nucleus provided with a karyosome. They have observed only one stage of the division of the nucleus, namely the formation of about a dozen plumous chromosomes from the karyosome, but they believe that it is a case of multiple nuclear division such as is known to occur in some of the gregarines and coccidia. After nuclear division the parts of the nuclear substance become scattered throughout the cytoplasm, but soon commence to arrange themselves side by side at the periphery. At this stage the parasite measures about 100 microns.

Up to this point in its development the parasite has one remarkable characteristic, namely, that its surface and the cyst membrane are not smooth but irregular. The cytoplasm becomes vacuolated and the nuclei project from the surface, and shortly afterwards are contained in little bud-like projections attached

to the general surface of the parasite by constricted necks. These buds become detached and rapidly increase in size until they measure 10 to 12 microns. These bodies are the sporoblasts.

The nucleus of each sporoblast divides into two and after an interval into a number of portions which arrange themselves in two rows or rather in two parallel planes, and segmentation occurs at right angles to these planes. The sporoblast is covered with a resistant membrane. It is these sporocysts which form the whitish powder in the body cavity of infected flies. Isolated sporozoites have been studied in preparations stained by Giemsa. They take the form of curved vermiform bodies. The nucleus is placed at about the posterior third of the body, which is rounded at the posterior and pointed at the anterior end. In front of and behind the nucleus there are two bodies which stain of a deep violet tint with Giemsa. The anterior one is by far the larger of the two.

The view that the parasite is a haemogregarine which passes through the stage of sporogony is justified by recent investigations into the life history of a number of haemogregarines. The parasite would appear to approach most nearly to *Hepatozoon perniciosum*.

The authors are inclined to think that the vertebrate host of the parasite is a crocodile or a lizard.

(430) JOUKOFF (N. M.). Culture du Parasite de la Malaria.—*Compt. Rend. Soc. Biol.* 1913. Jan. 24. Vol. 74. No. 3. pp. 136-138. With 1 text-figure.

The authors have succeeded in cultivating *Plasmodium praecox* and *Plasmodium malariae*. Their technique is as follows:—

Ten cubic centimetres of blood containing *Plasmodium praecox* were placed in each of a number of tubes and $\frac{1}{2}$ cc. of 10 per cent. sodium citrate solution was added. Some of the tubes were incubated at 41° and some at 44° C., the period of incubation in each case being half an hour.

As it was desired to produce the sporogonous forms the tubes so incubated were then incubated at 25° to 26°, since sporogony normally occurs in the mosquito at about this temperature.

Examinations were made every three hours.

No parasites could be found in the tubes which had been first incubated at 44° C. In the tubes incubated at 41° C. the first stages of the sexual cycle of the parasite were obtained. This culture survived only three days.

In the second experiment *Plasmodium malariae* was used. The tubes of citrated blood had added to them $1\frac{1}{2}$ cc. to 2 cc. of Lokke serum before incubation at 41° C. In these cultures the authors observed not only conjugation but also the subsequent segmentation of the fertilised elements. On the sixth day a large number of sporozoites of various shapes were seen, some in which division had not actually occurred, and some in which it was complete.

The segmentation was different from that observed in the mosquito, but this is due according to the authors to differences in the conditions compared with those prevailing in the mosquito.

- (431) ROWLEY-LAWSON (Mary). The Extracellular Relation of the Malarial Parasite to the Red Corpuscle, and its Method of securing attachment to the External Surface of the Red Corpuscle. —*Jl. Experiment. Med.* 1913. Mar. 1. Vol. 17. No. 3. pp. 324-343. With 6 plates.

In this paper the view is expressed that the malarial parasite is extra-corpuscular during the whole of its developmental cycle, and that it retains its hold upon the external surface of the red corpuscle by means of pseudopodia. Numerous photographs are given in the plates in support of this view.

HELMINTHS.

- (432) BRAU & BRUYANT (L.). Quelques Notes sur les Helminthes du Porc en Cochinchine. [Helminths occurring in the Pig in Cochinchina.]—*Bull. Soc. Path. Exot.* 1913. Jan. Vol. 6. No. 1. pp. 41-43.

Trematodes. In 1911 MATHIS and LEGER recorded the occurrence of *Fasciolopsis buski* in 6 per cent. of pigs at Tonkin; commenting at the same time on the rare occurrence of this parasite in man. They also drew attention to the statement of BARROIS and NOC that the parasite was of frequent occurrence in man in Cochinchina, and expressed the desirability of examining the pigs slaughtered at Saigon (Cochinchina).

The present authors made the desired examination of 100 pigs at Saigon, but were unable to find a single specimen of the parasite.

Although *F. buski* was not found, an amphistome possessing all the characters of *Gastrodiscus* Leuckart was found in 5 per cent. of the pigs examined. Save for minute differences in preserved specimens, which the authors think were due to shrinkage, the parasite was indistinguishable from *G. hominis*.

The discovery appears to confirm the idea that, although the parasite had been found only in man, it probably occurred also in an animal, and particularly in a domesticated animal.

Cestodes. *Taenia solium* would appear to be somewhat rare in Asia and particularly in Cochinchina, but the authors have received from DONNADIEU a pig's heart literally crammed with cysticerci, which on examination proved to be *C. cellulosae*.

Nematodes. Four species of nematodes have been encountered in the intestines of pigs:

Oesophagostoma dentatum in 95 per cent. of pigs.

Arduenna strongylina in 2 per cent.

Arduenna dentata in 2 per cent.

Trichuris crenatus in 3 per cent.

Only 5 per cent. of the pigs examined had no worm parasites in the alimentary canal.

- (433) MITTER (S. N.). Further Note on a *Gastrodisc*(?) from an Indian Zebu.—*Vet. Jl.* 1913. Mar. Vol. 69. No. 453. p. 117. With 1 text-figure.

The author has submitted specimens of this parasite, of which he has already published a description in the *Journal of Comparative Pathology and Therapeutics*, to RAILLIET for identification. The parasite is stated to be a *Homolostoger* [? *Homalogaster*], and the author believes that his specimens are a local variety of *H. paloniæ*. The figure given is an illustration taken from a paper by Benham in Lankester's "Zoology."

RABIES.

- (434) ACTON (H. W.) & HARVEY (W. F.). The Fixation of Rabies Virus in the Monkey (*Macacus rhesus*) with a Study of the Appearance of the Negri Bodies in the different Passages.—*Parasitology*. 1913. Jan. Vol. 5. No. 4. pp. 227-233. With 1 plate.

In the brief introductory portion of this paper reference is made to the opinion held by MAGENDIE that the rabies virus when transmitted by bite from dog to dog lost its virulence about the fifth passage. This view was also held by CELLI and MARINO-ZUCCO who considered that they had confirmed it by experiments.

In a recent experiment the authors have found that on the passage of a fixed virus of the rabbit through a dog the virus remained fixed for the dog, but in the fifth passage the animal did not show rabies. When this dog was re-inoculated 20 days later with material from the brain of the fourth dog in the series of passages it contracted dumb rabies after the fixed incubation period, thus showing that the loss of virulence was not due to attenuation, but to technique. They therefore hold that in all experimental work on rabies the controls should be equal in every respect to the test animals.

In *Macacus rhesus* the authors have found that the virus of rabies becomes fixed just as in the rabbit.

In the first two passages in this species the rabies was of the furious type; from the third to the sixth passage the symptoms commenced with fury, but for the last 24 to 48 hours there was complete paralysis. From the seventh passage onwards the paralytic type was alone observed.

In the monkey from the fourth passage onwards the incubation period varied from 7 to 8 days. The incubation period in rabbits inoculated from the monkeys showed a fixed virus period of incubation from the seventh passage onwards.

A converse experiment was also made to show that the fixed virus of the rabbit, when passed through *Macacus rhesus*, is not altered in its virulence either for this monkey or for the rabbit.

The formation of negri bodies.—

The results of the authors' study of Negri bodies in the ganglion cells of the hippocampus of *Macacus rhesus* have confirmed them in their view that they are nothing more or less than developments round nuclear fragments.

In each monkey a hundred ganglion cells were examined and the size, number, and colouration of the cell inclusions noted. The sections were stained with Mallory's iron haematoxylin and Bordeaux red.

The results of these examinations are summarised as follows:—

1. With subpassage, Negri bodies diminish in size until they become almost ultra-microscopic.

2. Of large size to begin with and staining brown to black, in the above experiment by the 6th passage they stain wholly black and are identical in staining reactions with the nucleolus.

3. There is a very decided drop in the numbers present at the 7th passage. The significance of this is not exactly clear. It is noteworthy, however, that the 7th passage is that in which the paralytic form of rabies is fully developed. In the 6th passage furious symptoms were still marked.

4. The diminution in the number of Negri bodies shown in the 7th passage is continued in the 8th and following passages.

5. The Negri bodies of the earlier passages (first three) showed a "veil" arrangement. Those of the later passages did not present this appearance, and may be described as "naked."

6. We regard the granules found in "veiled" Negri bodies as being composed of unaltered nucleolar substance.

7. The experiment given seems to show that the Negri body, as found in the cytoplasm of a ganglion cell, tends to disappear altogether from that region with the attainment of fixity of the virus. We have inoculated bullocks—the animals showing the largest Negri bodies known to us—one with street virus and another with fixed virus. The street virus brain showed large Negri bodies; the fixed virus brain showed none at all.

(435) WATSON (E. M.). *The Negri Bodies in Rabies.*—*Jl. Experiment. Med.* 1913. Jan. Vol. 17. No. 1. pp. 29-42. With 2 plates.

It would appear that a large proportion of the observations recorded in this paper were made on material taken from the brains of two dogs each of which died twenty days after having shown symptoms suspicious of rabies. The bodies of the dogs after having been burnt were exhumed for examination.

The author supports the view that Negri bodies represent the actual cause of rabies, and claims to have obtained evidence that they are of a protozoal nature. His conclusions are as follows:—

1. The Negri bodies, as the etiological agent in rabies, present two general types or phases in morphology, in growth, and in reproduction.

2. These two phases are constantly cyclic in their development and correspond (1) to a multiplicative, or schizogonous, and (2) to a reproductive, or sporogonous, life cycle.

3. By the detailed study of these forms and their succeeding stages we are inclined to believe that the Negri bodies are definite protozoan parasites, and from a study of their life history we are led to place them in the suborder of Cryptocystes, or Microsporidia, of the Sporozoa, and more definitely among the Oligosporogenea of the Glugeidae family, which forms produce but one pansporoblast.

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- (436) MACKIE (F. P.). *An Improved Method for Staining "Negri Bodies."*—*Indian Med. Gaz.* 1913. Jan. Vol. 48. No. 1. p. 20.

The following are the steps in the process:—

1. If the tissue has been fixed in Zenker's fluid the section must be treated with an alcoholic solution of iodine after the removal of the paraffin.

2. Wash with rectified spirit.

3. Flood the specimen with the following solution:

| | | | | |
|----------------------|-----|-----|-----|---------|
| Alcoholic eosin | ... | ... | ... | 0.5 g. |
| 50 per cent. alcohol | ... | ... | ... | 100 cc. |

4. Set fire to the solution on the slide and allow it to burn out, thus leaving a strong watery solution of the eosin on the section. This should be allowed to act for a minute or two.

5. Wash and run on a little rectified spirit.

6. Treat the section until it assumes a pink colour (a few seconds) with rectified spirit, to every cubic centimetre of which one drop of one per cent. caustic soda in alcohol has been added.

7. Wash in rectified spirit to every cubic centimetre of which one drop of five per cent. acetic acid has been added.

8. Stain with haemalum or haematoxylin.

9. Dehydrate, clarify and mount in balsam.

When sections are stained by this method the following result is obtained:—

The groundwork has a blue or pink tint; the nerve cells are blue; the nucleoli are red or violet; the red blood corpuscles bright red, and the Negri bodies vermilion or magenta.

The main idea of the method was obtained from MUIR.

The author suggests that the strong affinity possessed by Negri bodies for eosin indicates that the action of the dye upon the living virus might be tested. He has carried out a few inconclusive experiments himself.

UNDULANT FEVER.

- (437) RONCHÈSE (A.). *Sur le Séro-diagnostic de la Méliococcie avec des Cultures tuées par le Formol.* [The Serum Diagnosis of Undulant Fever with Cultures killed with Formalin.]—*Compt. Rend. Soc. Biol.* 1913. Feb. 7. Vol. 74. No. 5. pp. 210-212.

The experiments described in this paper were undertaken with the object of ascertaining whether cultures which have been preserved for a long period with formalin can be used for the agglutination test, or whether it is necessary to use freshly killed cultures.

The suspensions had added to them at the time of preparation two drops of formalin for every 15 cc.

In a series of tests the serum of persons who had recovered from the disease some months previously, and whose serum caused agglutination in a dilution of 1 in 100 was tested with (1) a fresh three-days-old culture of the micrococcus on agar, (2) an

emulsion made from a culture of the same age two months previously to which formalin had been added, (3) a similar suspension prepared a year previously.

It was found that the formalined suspensions were agglutinated to the same degree as the fresh one, and, further, that agglutination occurred more rapidly in the former than in the latter, the agglutination being the more rapid the older the emulsion.

Thus, the serum caused complete agglutination in the suspension of the fresh culture in dilutions of 1 in 10 and 1 in 50 in 40 minutes, and in a dilution of 1 in 100 in three hours. The periods necessary for complete agglutination with the emulsion prepared and formalined two months previously in the same dilutions were 15 minutes, 15 minutes, and 2 hours. With the emulsion prepared a year previously the periods were 5 minutes, 5 minutes, and 20 minutes respectively.

Tests were made with sera from people not infected with the organism, and it was found that there was no agglutination in dilutions of 1 in 10 even after three hours.

Heating the serum to 56° C. does not destroy its agglutinin either for the fresh emulsion or for formalined emulsions, but agglutination occurs a little more slowly.

(438) VALLET & RIMBAUD (L.). Étude expérimentale de l'Agglutination du *Micrococcus melitensis*.—*Compt. Rend. Soc. Biol.* 1913. Feb. 24. Vol. 74. No. 7. pp. 323-324.

In this paper are briefly summarised experiments carried out to investigate the following points:—

I. The agglutination of the micrococcus by normal sera.

II. The effect of other infections upon the agglutination.

III. The agglutination produced by the sera of immunised animals.

IV. Cross agglutinations with four different strains of the micrococcus.

I. The sera of healthy guinea-pigs as a rule agglutinate the organism in low dilutions only—1 in 20.

The serum of the rabbit while still possessing a feeble agglutinating power for the organism causes agglutination in slightly higher dilutions than guinea-pig serum.

The serum of normal dogs has a very high agglutinating power for the *Micrococcus melitensis*. The author tested the serum of 21 dogs and in 18 instances there was agglutination in dilutions varying from 1 in 20 to 1 in 400, and even 1 in 600.

Heating the serum to 56° C., as suggested by NEGRE and RAYNAUD, has constantly destroyed the agglutinating power.

The addition of fresh serum to heated serum does not restore the agglutinating power.

II. The sera of animals inoculated with *B. eberthi*, *B. coli*, staphylococci, and other organisms have been used, but in no case was any agglutination observed.

III. The intravenous inoculation of rabbits and dogs with cultures of the micrococcus killed by heat at 60° C. confers a

very high power of agglutinating upon the sera, or increases that already present to a marked extent. Positive results may be obtained in dilutions from 1 in 1,000 to 1 in 3,000.

In the rabbit and the guinea-pig subcutaneous and intraperitoneal inoculations have a less marked effect.

Heating the serum to 56° C. may cause complete disappearance of the agglutinin. It caused complete disappearance of the agglutinin in every one of the guinea-pigs, in one out of five rabbits, and in three out of four dogs, even when the sera caused agglutination in dilutions of 1 in 1,000.

In cases in which heating does not absolutely destroy the agglutinin the limit of positive reaction is very much lowered.

IV. The agglutinability of various strains with normal sera shows very great variation. Some sera which will agglutinate certain strains in dilutions up to 1 in 600 fail to produce any agglutination whatever with other strains.

The serum of an immunised animal as a rule agglutinates other strains, but this is not invariably the case.

In a single series of cross agglutination tests the strains which are most highly agglutinated are not of necessity those which have been used for the immunisation of the animals.

When heating does not destroy the agglutinating power the agglutination indexes of the different strains before and after heating are not proportional.

MISCELLANEOUS.

- (439) VON PROWAZEK (S.). Untersuchungen über die Tona der Pferde auf Samoa. [Tona in Horses in Samoa.]—*Arch. f. Schiffs- u. Trop.-Hyg.* 1913. Jan. Vol. 17. No. 1. pp. 1-8. With 4 text-figures.

In Samoa horses are frequently the subject of granulating ulcers below the eyes and somewhat large granulating tumours which for the most part occur in the region of the fetlock and to which the term "Tona" is applied. The same term is used to designate framboesia in man.

KRAMER and HENGELER, and FRANKE do not think that there is any connection between the disease in horses and the condition affecting the human subject. The affection of the horse results, in their view, from the neglect of wounds caused by thorns.

The present author states that the two diseased conditions are in reality quite distinct. In primary lesions in man typical framboesia spirochaetes can be demonstrated, but this is not the case with the equine disease.

The disease occurring in the neighbourhood of the eye differs from that involving the leg in that it usually takes the form of a deep ulcer not unlike "ulcus tropicum." The ulcers are crater-like and covered with a dirty brown, grey, or blood-stained material.

In sections pyogenic cocci are principally found, and the lesion is due to ordinary injuries in which the healing processes are delayed.

The lesion occurring on the leg is, however, of quite a different nature. This takes the form of a granulating tumour varying in size up to that of an apple and in many cases presenting a papillomatus appearance.

Microscopic examination shows that in these lesions there is marked development of the papillae of the cutis, associated with hyperkeratosis. In the basal layer of the epithelium there is active proliferation. In many cases a secondary lesion in the form of a crop of pustules occurs around the periphery of the growth. In the superficial parts numbers of bacteria and cocci can be seen, but in the deeper layers fusiform bacilli and, still deeper, large numbers of spirochaetes, some of which actually appear to be intracellular, can be detected.

The author refers at some length to the various systems of nomenclature that have been adopted by different authors for the designation of the parasites generally referred to under the name "spirochaetes" and concludes that the most correct name is "*Borrelia*," although the most descriptive is "*Spirosoma*" (Schilling).

Since in the group of skin diseases at least three types of *Borreliae* occur, viz., large, intermediate, and small he suggests that the three forms occurring in "Tona" in the horse should be termed *B. tonae magna*, *media*, and *minima*.

Many observers consider that "tona" in the horse is due to systemic disease, while others consider it to be a localised disease of the foot which disappears when the animal is removed to a higher locality. This rather connects the condition with injuries caused by thorns, etc.

The author failed to transmit the condition experimentally in one instance.

In the deep layers of the epithelium eosinophilic cell-inclusions can be seen and these may be considered as parasites, but they are very scanty and the granules may be looked upon as nuclear fragments. The spirochaetes play a secondary part, penetrating into the deeper layers, while the bacteria remain in the more superficial portions of the growth. The spirochaetes may convert the tumour-like growth into a lesion resembling *ulcus tropicum*.

The spirochaetes may be made to disappear by frequent painting of the lesion with 20 per cent. eosin, the animal being kept in the sun. Smearing with precipitate ointment or painting with silver nitrate has the same effect.

(440) CARTER (H. F.) & BLACKLOCK (B.). External Myiasis in a Monkey.—*Brit. Med. J.* 1913. Jan. 11. p. 72.

The authors observed two small groups of dipterous larvae upon a monkey (*Cercopithecus callitrichus*) which was the subject of acute tuberculosis and died on the day after the observation was made. One batch was found in the nostril and on the side of the face, and the other on the side of the body near the groin.

In both batches there were larvae at different stages of development indicating that they had not all been deposited at the same time. In all 21 larvae were removed. Sixteen of them were of the muscid type and the remainder were specimens of *Fannia canicularis*. From the muscid larvae *Calliphora erythrocephala* and *Muscina stabulans* were obtained.

- (441) VON RATZ (S.). Fütterungsversuche mit dem Virus der infektiöse Bulbärparalyse. [Feeding Experiments with the Virus of Infectious Bulbar Paralysis.]—*Zeitschr. f. Infektionskrankh. Parasit. Krankh. u. Hygiene d. Haust.* 1913. Vol. 13. No. 1-2. pp. 1-7.

In the first series of experiments, brief details of which are given, nine mice were fed with brain tissue derived from cats or rabbits dead of the disease. Six of these animals became infected and died, death taking place on the 2nd to the 15th day. Nervous tissue from three of these mice was used for the inoculation of control rabbits, two of which died with typical symptoms.

In mice no characteristic symptoms are observed, but there was some paralysis of the hind legs.

Positive results were obtained with two white rats, the animals dying on the 17th and 18th days, and rabbits were successfully inoculated from them.

Feeding experiments with rabbits and guinea-pigs failed even though some of the rabbits were first treated with a 1 per cent. solution of sulphuric acid in order to produce intestinal catarrh.

Of 11 cats and dogs fed with infective material five became infected, and the symptoms produced correspond exactly with those observed in natural cases. As in the experiments with rabbits, some of the cats were previously given dilute sulphuric acid.

The author states that his experiments do not furnish absolute proof that the virus can be absorbed through sound mucous membranes, but he believes that such absorption is possible and quotes in support of this view the successful experiments of GALTIER, HOGYES, REMLINGER, and CONTE with the virus of rabies.

- (442) BALFOUR (A.). Nasal Lesions in Glanders and Epizootic Lymphangitis.—*Bull. Soc. Path. Exot.* 1913. Mar. Vol. 6. No. 3. pp. 145-146.

After referring to BRIDGÉ's report of the occurrence of nasal lesions in these diseases, the author states that he saw a mule at Mongalla which presented symptoms suggestive of glanders. There was a large swelling in the nasal region which was discharging thick pus, and there was a purulent discharge from the nose. There was no glandular enlargement. Microscopic examination of the pus revealed the presence of the *Saccharomyces farciminosus* in large numbers. Cultures showed no evidence of the presence of *B. mallei*. No mallein was available. On examining the skull it was found that there had been extensive ulceration resulting in absorption of a large portion of the malar bone.

- (443) PETTIT (A.). Procédé simple pour prélever du Sang chez les petits Rongeurs. [Simple Method of obtaining Blood from small Rodents.]—*Compt. Rend. Soc. Biol.* 1913. Jan. 10. Vol. 74. No. 1. pp. 11-12.

The method described in this paper is said to be equally applicable to the guinea-pig, rat, and mouse.

A glass pipette which is drawn out to a moderately fine point is inserted at the inner canthus between the eye-ball and the wall of the orbit. It is passed along the wall of the orbit from before backwards, keeping it inclined at an angle of about 45 degrees, and directing it towards the optic nerve so as to perforate the cavernous sinus.

In the mouse the pipette requires to be passed in for a distance of about 5 millimetres, in the rat 10, and in the guinea-pig about 12.

As a rule the pressure of the blood is sufficient to force it up the pipette, but slight suction may be exerted by means of a rubber teat.

In order to lessen the risk of contamination of the blood some drops of an antiseptic may be instilled into the eye.

The method may be used when it is required to make intravenous inoculations into these animals.

RECENT LITERATURE.

[Continued from BULLETIN No. 3, pp. 193-196.]

Foot-and-Mouth Disease.

- (444) BÖHM (J.). Zur Pathogenese der Maul- und Klauenseuche. [The Pathogenesis of Foot-and-Mouth Disease.]—*Zeitschr. f. Fleisch- und Milchhygiene*, 1913. Mar. 15. Vol. 23. No. 12, pp. 266-267.
- (445) LOEFFLER (F.). Versuche über die Abtötung des Ansteckungstoffes der Maul- und Klauenseuche in vorschriftsmässig gepacktem Dünger. [Experiments in connection with the Destruction of the Virus of Foot-and-Mouth Disease in Dung.]—*Berl. Tierärzt Wochenschr.*, 1913. Feb. 13. Vol. 29, No. 7, pp. 113-115.

Leishmaniasis.

- (446) GABBI (U.). Au Sujet de l'Histoire du Kala-azar Méditerranéen.—*Bull. Soc. Path. Exot.*, 1913. Mar. Vol. 6. No. 3, pp. 141-143.
- (447) LAVERAN (A.). Au Sujet de l'Histoire du Kala-azar Méditerranéen.—*Bull. Soc. Path. Exot.*, 1913. Jan. Vol. 6. No. 1, pp. 23-24.
- (448) MIGONE (L. E.). Un Cas de Kala-azar à Asuncion (Paraguay).—*Bull. Soc. Path. Exot.*, 1913. Feb. Vol. 6. No. 2, pp. 118-120.

Malaria.

- (449) ZIEMANN (H.). Ueber die künstliche Weiterentwicklung (in vitro) des Tertian-Malariaparasiten. [The Artificial Cultivation of the Parasite of Tertian Malaria in vitro.]—*Deut. Med. Wochenschr.*, 1913. Feb. 6. Vol. 39. No. 6, p. 260.

Plague.

- (450) GALLI-VALERIO (B.). *Bacterium pseudopestis murium* n. sp.—*Centralbl. f. Bakt.*, 1. Abt., Orig., 1913. Mar. 1. Vol. 68. No. 2, pp. 188-194. With 5 text-figures.

Rabies.

- (451) MIESZNER (H.), KLIEM, & KAPFERGER. Immunisierungsversuche gegen Tollwut. [Immunisation against Rabies.]—*Arch. f. Wissenschaft. u. Praktisch. Tierheilkunde*, 1913. Feb. Vol. 39. No. 3, pp. 169-209.

Spirochaetosis.

- (452) NUTTALL (G. H. F.). The Herter Lectures. I. Spirochaetosis. Lecture delivered on the Herter Foundation, Johns Hopkins University, Baltimore, Maryland, U.S.A., Oct. 8, 1912.—*Parasitology*, 1913. Jan. Vol. 5. No. 4, pp. 262-274.

Trypanosomiasis.

- (453) BRUMPT (E.). Evolution de *Trypanosoma lewisi*, *duttoni*, *nabiasi*, *blanchardi*, chez les Puces et les Punaises. Transmission par les Déjections. Comparaison avec *T. Cruzi*. [*T. lewisi*, *duttoni*, *nabiasi*, *blanchardi* in Fleas and Bugs. Transmission by means of Dejecta. Comparison with *T. Cruzi*.]—*Bull. Soc. Path. Exot.*, 1913. Mar. Vol. 6. No. 3, pp. 167-171.
- (454) ——. Immunité partielle dans les Infections à *Trypanosoma cruzi*, Transmission de ce Trypanosome par *Cimex rotundatus*. Rôle régulateur des Hotes intermédiaires. Passage à travers le Peau.—*Bull. Soc. Path. Exot.*, 1913. Mar. Vol. 6. No. 3, pp. 172-176.

- (455) CHATTON (E.). Position Systématique et Signification Phylogénique des Trypanosomes Malpighiens des Muscides. Le genre *Rhynchoidomonas* Patton.—*Compt. Rend. Soc. Biol.*, 1913. Mar. 14. Vol. 74. No. 10, pp. 551-553.
- (456) — & LEGER (M.). L'Autonomie des Trypanosomes propres aux Muscides démontrée par les élevages purs indéfinis.—*Compt. Rend. Soc. Biol.*, 1913. Mar. 14. Vol. 74. No. 10, pp. 549-551.
- (457) LAZILLO (V.). La Durina (Sifilide Equina) in Due Asini. [Dourine in two Asses.].—*Giorn. di Med. Vet.*, 1913. Jan. 18. Vol. 62. No. 3, pp. 45-49.
- (458) NUTTALL (G. H. F.). The Herter Lectures. II. Trypanosomiasis. Lecture delivered on the Herter Foundation, Johns Hopkins University, Baltimore, Maryland, U.S.A., Oct. 9, 1912.—*Parasitology*, 1913. Jan. Vol. 5. No. 4, pp. 275-288.
- (459) RANKEN (H. S.). A Preliminary Report on the Treatment of Human Trypanosomiasis and Yaws with Metallic Antimony.—*Proc. Roy. Soc.*, 1913. Mar. 5. Series B. Vol. 86. No. B 586, pp. 203-215.
- (460) RINGENBACH (J.). Contribution à l'Etude de la Distribution de la Maladie du Sommeil en Afrique équatoriale française (Pays Bakongo, Bakongui et Loango) Mai-Juin-Juillet 1912.—[The Distribution of Sleeping Sickness in Equatorial French Africa. May-June-July, 1912.].—*Bull. Soc. Path. Exot.*, 1913. Jan. Vol. 6. No. 1, pp. 34-40.
- (461) RODHAIN (J.). A propos de *Leptomonas pangoniæ* et *Trypanosoma denysi*. Note rectificative.—*Bull. Soc. Path. Exot.*, 1913. Mar. Vol. 6. No. 3, pp. 181-182.
- (462) UHLENHUTH (P.), MULZER (P.) & HÜGEL (G.). Die chemotherapeutische Wirkung von organischen Antimonpräparaten bei Spirochäten- und Trypanosomenkrankheiten. [The Actions of Organic Compounds of Antimony in Spirochaetosis and Trypanosomiasis.].—*Deut. Med. Wochenschr.*, 1913. Feb. 27. Vol. 39. No. 9, pp. 393-395.

Biting Flies and Ticks.

- (463) CHRISTOPHERS (S. R.). Contributions to the Study of Colour Marking and other Variable Characters of Anophelinae with special reference to the Systematic and Phylogenetic Grouping of the Species.—*Ann Trop. Med. & Parasit.*, 1913. Mar. 31. Vol. 7. No. 1, pp. 45-100. With 4 plates.
- (464) HADWEN (S.). The Life-history of *Dermacentor Variabilis*.—*Parasitology*, 1913. Jan. Vol. 5. No. 4, pp. 234-237.
- (465) NEWSTEAD (R.). Phlebotomus from West Africa.—*Bull. Soc. Path. Exot.*, 1913. Feb. Vol. 6. No. 2, pp. 124-126.
- (466) ROUBAUD (E.). Quelques Mots sur les Phlébotomes de l'Afrique occidentale française. [Notes on the Phlebotomus of French West Africa.].—*Bull. Soc. Path. Exot.*, 1913. Feb. Vol. 6. No. 2, pp. 126-128.

Helminthiasis.

- (467) MIYAGAWA (Y.). Ueber den Wanderungsweg des *Schistosomum japonicum* durch Vermittlung des Lymphgefäßsystems des Wirtes. II. Mitteilung. [The Path taken by *Schistosomum japonicum* in the Lymphatic System of the Host.].—*Centralbl. f. Bakt.*, 1. Abt., Orig., 1913. Mar. 1. Vol. 68. No. 2, pp. 204-206.
- (468) —. Ueber den Wanderungsweg des *Ankylostomum duodenale* (*caninum*) bei Oraler Infektion. Vorläufige Mitteilung. [The Path followed by *Ankylostomum duodenale* (*caninum*) following Ingestion of the Parasite.].—*Centralbl. f. Bakt.*, 1. Abt., Orig., 1913. Mar. 1. Vol. 68. No. 2, pp. 201-204.

Protozoa.

- (469) BERGMAN (A. M.). Beitrag zur Kenntnis des Vorkommens der Sarkosporidien bei der Haustieren. [The Occurrence of Sarcosporidia in the Domesticated Animals.]—*Zeitschr. f. Fleisch- und Milchhygiene*, 1913. Jan. 15. Vol. 23. No. 8, pp. 169-180.
- (470) MARULAZ (M.) & ROUDSKY (D.). Contribution a l'Étude de *Haemogregarina terzii* Sambon et Seligmann.—*Compt. Rend. Soc. Biol.*, 1913. Jan. 24. Vol. 74. No. 3, pp. 128-131. With 9 text-figures.
- (471) SPLENDORE (A.). Sulla Toxoplasmosi dei Conigli. [Toxoplasmosis of the Rabbit.]—*Pathologica*, 1913. Jan. 15. Vol. 5. No. 101, pp. 48-52.

Unclassed.

- (472) BAUJEAN (R.). Note sur le Venin de *Bitis arietans* ou Vipère heurtante. [The Venom of the Puff Adder.]—*Bull. Soc. Path. Exot.*, 1913. Jan. Vol. 6. No. 1, pp. 50-54.
- (473) MOHLER (J. R.) & EICHHORN (A.). Immunisation against Haemorrhagic Septicaemia.—*American Jl. Vet. Med.*, 1913. Jan. Vol. 8. No. 1, pp. 14-19.
- (474) MORETTI (E.). Contributo all' Immunizzazione contro il Barbone bufalino. [Immunisation against Barbone.]—*Jl. Med. Zootatro. Parte Scientifica*, 1913. Feb. Vol. 24. No. 2, pp. 60-74.
- (475) MROWKA. Unsere Haustiere in Ostasien, ihre Eigenart und ihre Krankheiten mit Berücksichtigung der Parasiten. [Our Domesticated Animals in Eastern Asia, and their Diseases with Particular Reference to Parasites.]—*Zeitschr. f. Veterinärkunde*, 1913. Mar. Vol. 25. No. 3, pp. 97-107. With 5 figures.
- (476) PHISALIX (Marie). Propriétés Vaccinantes du Venin muqueux de la Peau des Batraciens contre lui-même et contre le Venin de la Vipère Aspic. [The Protective Properties of the Mucous Poison of the Skin of the Batrachians against Itself and against the Poison of the "Aspic" viper.]—*Bull. Soc. Path. Exot.*, 1913. Mar. Vol. 6. No. 3, pp. 190-195.
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TROPICAL DISEASES BUREAU.

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[Vol. 1.

BABESIASIS (PIROPLASMOSIS) AND ANAPLASMOSIS.

- (477) SCHELLHAASE (W.). Beobachtungen über die Anaplasmosis und Piroplasmosis der Schafe und Ziegen in Deutsch-Ostafrika. [The Occurrence of Anaplasms and Piroplasms in the Sheep and Goat in German East Africa.]—*Zeitschr. f. Infektionskrankh. Parasit. Krankh. u. Hyg. d. Haust.* 1913. June. Vol. 13. No. 6. pp. 349-352.

In a previous paper (see this *Bulletin*, Vol. 1, No. 2. Abstract No. 117) the author recorded the occurrence of anaplasms in two sheep.

In the present paper he records the presence of similar bodies in the blood of two further sheep. The animals were in poor condition and the bodies were far more scanty in the blood than is observed in cattle infected with the parasite. In addition to these four animals the author has found "marginal points" in very small numbers in another batch of sheep. All the animals were in poor condition and this was the only constant symptom. In some of the cases other symptoms such as diarrhoea, rhinitis, conjunctivitis and shedding of the wool were observed, but these symptoms were not constant. Occasional "marginal points" were seen in the blood of 5 out of 18 diseased sheep.

In examining the blood of diseased sheep the author has found piroplasms resembling *P. mutans* in 6 out of 16 animals. These animals showed the same symptoms as those in which the anaplasms were found. In four of these instances both piroplasms and anaplasms were found in the same animal.

The author has also found in a few goats that were emaciated very small numbers of bodies resembling "marginal points" and piroplasms.

- (478) BEVAN (Ll. E. W.). Some Observations on the different Strains of Bovine Plasmoses in South Africa and the Immunity conferred by them in Southern Rhodesia.—*Veterinary Jl.* 1913. May. Vol. 69. No. 455. pp. 208-212.

In 1911, 48 animals received from Great Britain were inoculated with virus supplied by THEILER which contained *B. bigemina* and *A. centrale*. None of these animals died during

the process of immunisation and reports were received regarding 38 of them after distribution. Of those reported upon 11 died and 8 suffered relapses with recovery.

During the same period 10 animals which had been inoculated at Pretoria were also inoculated with the local virus. Of these two died during the immunisation. Reports were received regarding five of the animals distributed. None of these died, and none had relapses.

These figures appear to show that the Pretoria virus does not confer complete immunity against the local virus. This was to some extent foreseen inasmuch as THEILER had found that "a recovery from infection with *A. centrale* does not cause complete immunity."

It was found that although the animals inoculated at Pretoria showed marked reactions, the virus had become markedly attenuated if not inert.

It having been found that the Pretoria virus did not confer complete immunity upon imported cattle exposed to natural infection in Southern Rhodesia, it was considered advisable again to make use of an attenuated local virus.

It was considered better that the animals should suffer from the infection while under veterinary supervision than unexpectedly after distribution.

During the year 1912-1913 seventy-seven animals from the Southern Colonies and sixty-eight from Great Britain were successfully immunised. Twenty-five home-bred animals died, but the animals were not in a suitable condition for immunising and their owners had been warned that accidents might occur.

Twenty-five animals which had been inoculated with redwater in Great Britain before shipment, and which had had more or less severe reactions, were inoculated on arrival with mixed babesiasis and anaplasmosis virus (Rhodesian), and all except two reacted to both of the infections.

Disappointing results have attended the use of trypanblue which, while controlling redwater, appears to produce a condition unfavourable to recovery from the anaplasmosis which follows. It is especially harmful in pregnant animals.

(479) CRUCA (A.). A propos de l'Immunité Active du Chien vis-à-vis de la Piroplasmose Canine (Babesiose canine).—*Bull. Soc. Path. Exot.* 1913. July. Vol. 6. No. 7. pp. 499-501.

A dog three months old was inoculated intraperitoneally with 16 cc. of blood containing a large number of organisms, the strain being the Tonkin virus of MATHIS. Three days later there was an elevation of temperature, and parasites appeared in the blood on the fifth day. The disease was acute and for eight days the blood was very rich in parasites. After this, parasites disappeared from the circulation and the dog recovered.

Twenty-six days after the disappearance of the parasites the blood was used for the inoculation of another dog with positive result, the dog dying on the 10th day.

Nine months after the disappearance of parasites from the blood a second dog was inoculated intraperitoneally; an acute attack was set up but the dog did not die.

Twenty-eight months after the disappearance of the parasites from the blood the intraperitoneal inoculation of 20 cc. of blood into another dog failed to produce any symptoms.

The original dog which had recovered from the acute attack following the inoculation with the Tonkin virus was then inoculated intraperitoneally with 5 cc. of defibrinated blood obtained from NUTTALL. Two days later there was a rise of temperature and parasites appeared in the blood. There was a rapid increase in the number of parasites present in the blood, jaundice and haemoglobinuria appeared, and the dog died on the 13th day after inoculation.

The author concludes that when a dog has made a complete recovery it is, at least in some cases, susceptible to fresh infection. The author "admits the identity of the parasites of MATHIS (Tonkin) and that of NUTTALL (South Africa)."

LAVERAN pointed out that dogs immune to a virus obtained from one source are not always immune to viruses obtained from other sources.

(480) ZIEMANN (H.). Über die Kultur der Malariaparasiten und der Piroplasmen (*Piroplasma canis*) in vitro. [The Cultivation of the Malaria Parasites and Piroplasms (*Piroplasma canis*) in vitro.]—*Arch. f. Schiff- u. Trop.-Hyg.* 1913. June. Vol. 17. No. 11. pp. 361-391. With 2 coloured plates.

In this abstract the cultivation of *Piroplasma canis* only is dealt with.

After many preliminary experiments the author found that it was inadvisable to obtain blood for cultures from animals that were severely infected and in whose blood parasites were numerous. The best results are obtained with blood in which the parasite has just made its appearance, and before any serious blood alterations have taken place. If dogs of more than 3 or 4 months old are alone available, the author advises that splenectomy should be performed and that about four days after they should be bled to the extent of 30 to 50 cc. of blood according to the size of the animal in order to produce a marked anaemia before they are infected. A large number of media were tried, but the best results were obtained in the following manner:

A young healthy dog is bled from the carotid into a glass cylinder with a narrow neck. The blood is carefully defibrinated, care being taken to avoid the formation of bubbles. After removal of the fibrin, 50 per cent. solution of dextrose is added in the proportion of 0.2 cc. to 10 cc. of the defibrinated blood, and in addition to this 0.3 cc. of a 2 per cent. sodium citrate in 0.85 per cent. salt solution is added to every 10 cc. of defibrinate blood. In a number of cases the sodium citrate solution did not appear to be so necessary.

The mixture is centrifuged and leucocyte-free blood is pipetted from the bottom and placed in tubes which have been filled to a height of at least 5 centimetres with inactivated (sodium citrate)-dextrose-dog-serum or (sodium citrate)-dextrose-ascitic fluid. In order to be able to work quickly fresh normal dextrose-dog-serum or dextrose-human-ascitic-fluid should be inactivated before the infected blood is withdrawn from the dog.

The author was able to obtain cultures although leucocytes were present. Inactivated human-dextrose-serum was found in some cases to be as good for cultivation purposes as the substances already mentioned. Horse serum was the worst.

The layer of corpuscles transferred to the culture tubes should not be too thin. Cultures can be obtained at 40° C. and at room temperature, but the best results were obtained at 37° C.

It is remarkable to what extent multiplication occurs within 24 hours. While dividing forms with more than four merozoites are seldom met with in the peripheral blood of dogs, in cultures there are frequently found schizonts with 16 or more merozoites. If it is desired to preserve the parasites without marked multiplication, it appears to be best to keep the tubes at room temperature. The addition of a greater proportion of dextrose does not appear to make any great difference.

The author was able to keep cultures alive for 5 or 6 days, and in one instance for 16 days. By the second day exhaustion of the medium or the formation of a toxin leads to degeneration of some of the parasites, and in any case the multiplication is most rapid during the first day.

The author succeeded in infecting dogs by intravenous inoculation with subcultures four or five days old, and in one instance he was able to obtain subcultures from a culture that was four days old. From these results it would appear that the virulence of the organism is retained in cultures. In tubes that become contaminated with bacteria the parasites rapidly die.

When degeneration occurs the parasites lose their typical morphology and become rounded. The cytoplasm stains more intensely and the chromatin less intensely. The cytoplasm may simultaneously break up into fragments.

No indication of sporogony is obtained in cultures kept at room temperature.

In two instances dogs were inoculated subcutaneously with dead cultures, but no protection against living cultures was conferred upon them.

In one instance no multiplication took place when blood from an animal that was the subject of acute piroplasmosis was added to blood taken from an apparently healthy dog which was immune to the parasite, while growth took place in control tubes containing blood from healthy dogs. As in the case of the malaria parasite, not every blood appears to be suitable for the cultivation of the parasite.

It is worthy of note that the serum of the immune dog (not inactivated) appeared to have a more rapid destructive effect upon the parasite than uninactivated serum from a normal dog.

- (481) DONN (S.). Anaplasms or Jolly Bodies? A Contribution to the Knowledge of Certain Intracorpuseular Bodies present in the Blood of Some Species of Mammals.—*Jl. Comp. Path. & Therap.* 1913. June. Vol. 26. No. 2. pp. 97-110. With 6 figures.

In this article are described bodies presenting morphological and staining characters identical with or very similar to those possessed by anaplasms. They have been observed in the blood of wild animals such as the lemur, mouse deer, orang-utang, capuchin monkey, etc.

Up to the present no convincing evidence has been brought forward to show whether the chromatin bodies discovered in the blood of animals, other than cattle or rodents, should be classified with the anaplasmata of THEILER or with the bodies of JOLLY who first described their occurrence in the latter type of animal.

In view of the various opinions expressed by a number of authors regarding the occurrence and the nature of bodies of this kind occurring in different species, the author records the results of observations made upon nearly 300 animals of a variety of orders, excluding the domestic species.

Chromatin bodies have been observed in the mouse deer (*Tragulus javanicus*). In this animal the red corpuscles measure only 2.5 microns. The chromatin-staining bodies were found either in the marginal or in the central position, and they varied a little in size. The bodies were relatively numerous, there being one or more in every field, but no double ones were demonstrated. These bodies were only found in the blood of one out of three mouse deer examined.

Bodies possessing the same characters were found in the blood of five lemurs which died, some after having shown signs of illness and others without any illness having been observed. The marginal disposition of the bodies was noted. On account of this, blood examinations were made of eight lemurs belonging to four different species and chromatin bodies were found in variable numbers in all.

Out of 25 of the order *Quadrumania* examined by the author during the last two years chromatin bodies were found in the blood of two only.

In a capuchin monkey which had died as a result of the formation of metastatic abscesses the bodies were numerous present, but the disposition of them within the corpuscles was indifferent. There was considerable variation in size of the bodies, and none of them were seen in an extracorpuseular position. These points contra-indicate the view that they were in reality staphylococci.

In an orang-utang, the primary cause of whose death was infection with *Haemoproteus pitheci*, chromatin bodies were found in small numbers in the corpuscles from the general circulation and from the organs, but in smears made from the marrow of the long bones they were relatively numerous. In a number of the erythroblasts present in these smears the nuclei were undergoing fragmentation, and in a single smear all stages of fragmentation could be observed from the intact nucleus down to small spherical

fragments indistinguishable from the chromatin bodies. Further, in some of the corpuscles round nuclear fragments of the shape and size of the chromatin bodies could be seen just about to be detached from the parent nucleus. Cells containing nothing but small rounded bodies like those in the general circulation were common. Occasionally small dividing forms about the size of a coccus or less could be seen.

In smears from the marrow of the ribs of a hog deer (*Cervus porcinus*) a number of chromatin bodies were found in the red cells. The size of the bodies varied more considerably than those seen in the general circulation of other animals. This deer had been in the Zoological Gardens for a number of years, and up to the time of death no marked illness had been observed.

The author has observed bodies of this kind in the blood of very nearly every marsupial animal that he has examined. He has found them in animals in the best of health, wild and captive, adult and immature. Owing to this it is practically impossible to carry out any inoculation experiments. Although normally present in the blood it is legitimate to consider that their number may be increased in certain pathological conditions, such as anaemia due to stomach worms, etc. As in other species the bodies vary greatly in size.

The author's findings in the examination of the blood of a number of lemurs were practically the same as in the case of the marsupials, but as the lemur is much higher in the zoological scale the author is not prepared to admit that the conclusions drawn with regard to the bodies in the blood corpuscles of the marsupials are also to be drawn regarding the bodies found in the lemurs, although it is pointed out that according to MORRIS such bodies may be found in the blood of normal cats.

The chromatin bodies were present in the blood of one mouse deer only out of three that were examined, and the blood of those in which the examination was negative was examined both before and after death. It is pointed out that the red corpuscles of the mouse deer are only about one-third the size of those of the other species examined and that in them the bodies were correspondingly small. It is suggested that this is further evidence tending to show that they are in reality remains of nuclei, as it would be extraordinary to find a smaller parasite choosing for its host an animal with a smaller red corpuscle.

THEILERIASIS.

(482) THEILER (A.). The Immunisation of Cattle against East Coast Fever.—*Second Report of the Director of Veterinary Research. Union of South Africa. October 1912.* pp. 266-314. (1913. Cape Town: Cape Times, Ltd., Government Printers.)

In the First Report of the Director of Veterinary Research it was recorded that out of 170 animals inoculated at the laboratory in various ways, 84 survived injection and natural infection. The method was introduced on a small scale in practice and since then has been considerably extended.

Up to September 1912, 133,833 cattle had been inoculated in the Transkei with the object of protecting as many of the animals as possible, since dipping could not be resorted to.

While the work was being carried on in the field, further investigations were made in the laboratory with the object of conferring a greater immunity by inoculation. In view of the fact that quinine is a plasma poison the effect of soaking the pulp used for the inoculations in varying strengths of quinine solution for half an hour before use was investigated.

Different grains of pulp were used, as follows:—

Fine grain.—A Latapie apparatus was used for the preparation of this.

Half Medium, Medium grain.—The pulp was minced once in a "Spong" apparatus, No. 42, medium size 12 teeth, to give the medium grain, and to obtain the half-medium grain it was put through the mincer twice.

Coarse and Half-Coarse grain.—An ordinary mincing machine was used, the pulp being put through once or twice respectively.

Part I.

A. *The immunisation of cattle obtained from non-infected areas.*

Experiment 1.—Nineteen animals were injected intravenously with coarse grain spleen and gland pulp taken from an animal slaughtered in the last stages of the disease and were immediately exposed to natural infection. Of these 9 survived, *i.e.*, 47.4 per cent.

Experiment 2.—Forty animals were injected into the jugular vein with 5 cc. of coarse grain spleen and gland pulp mixed with peptone. Twelve died of the injection, 25 reacted, and 3 failed to react. Of the 28 survivors, two died of the disease when exposed to natural infection. Percentage of survivors, 65.

Experiment 3.—Forty-eight animals were injected with 5 cc. of spleen and gland pulp of various grains and exposed to natural infection 13 days later. Twenty-two or 45.8 per cent. survived.

Experiment 4.—In this experiment the conditions of the preceding experiment were repeated, save that the animals were kept for 15 days before being exposed to natural infection. Forty-two animals were used, and of these 64.3 per cent. survived.

Conclusion.—For the inoculation of cattle in a non-infected area, the experience in the field indicates that the best results are to be obtained by the injection of 5 cc. of spleen or gland pulp (medium or half coarse grain), mixed with peptone, the injected animals to be exposed to veld infection 14 to 15 days later.

B. *The immunisation of cattle in infected areas.*

Experiment 5.—To note the effect of the intravenous injection of 5 cc. spleen or gland pulp, medium grain, mixed with peptone, the animals being placed upon infected veld immediately afterwards. Twenty-four animals were used. Of these 10 survived. Of the 14 that died, four were infected previous to inoculation

and 5 died as the result of being worked while passing through the reaction. Excluding these five, the survivors numbered 52·6 per cent. The animals that were infected before inoculation are included because this is one of the factors to be expected when animals are on infected veld.

Experiment 6.—To test the effect of intravenous injections of 10 cc. of pulp mixed with peptone and aleuronat. Of 43 injected, 22 or 51·2 per cent. survived. These animals were exposed to natural infection immediately after injection.

Experiment 7.—Thirty-one animals were injected intravenously with 10 cc. of pulp mixed with peptone, and immediately afterwards turned out on infected veld. 45·2 per cent. survived.

Experiment 8.—The injection of 10 cc. of pulp mixed with aleuronat, the animals being protected from natural infection till the 10th day. Of 30 animals used in the experiment, 14, or 46·7 per cent. survived.

Experiment 9.—The intra-jugular injection of 10 cc. of coarse grain pulp, the animals being protected from natural infection till the 10th day. Forty-eight animals were used, and of these 45·8 per cent. survived. Four of these animals contracted the disease and recovered.

Experiment 10.—In this experiment the conditions of the previous experiment were repeated save that the pulp was mixed with aleuronat. Of 64 animals used, 46, or 71·9 per cent. survived.

Experiment 11.—The conditions in this experiment were the same as in Experiment 9 save that the animals were kept on a clean farm until the 14th day. Seventeen animals were used, and of these 6 survived.

Experiment 12.—In this the conditions of Experiment 10 were reproduced except that the animals were protected from natural infection till the 14th day. Thirty-three animals were used. Seven died of the disease or as a result of the injection, 2 were drowned and are therefore excluded, six died of other causes, and of the surviving 18, one died of the disease on the 62nd day. The survivors were therefore 17 out of 31, or 54·8 per cent.

Experiment 13.—The injection of 5 cc. of coarse grain pulp mixed with peptone, the animals being kept on clean ground till the 14th day. In this experiment there were 154 survivors out of 266 animals, or 57·9 per cent.

Experiment 14.—In this experiment medium and coarse grain pulp were used, mixed with peptone, or peptone and aleuronat. The injected animals were kept in a clean paddock till the 14th day. Of 109 animals injected, 72, or 66 per cent. survived.

Experiment 15.—In this experiment the dose of pulp used was 10 cc. and it was mixed with peptone. The animals were protected from infection for 14 days as before. Of the 142 animals used, 97, or 68·3 per cent. survived.

The conclusion drawn from experiments 5 to 15 is as follows:—

For the inoculation of infected herds the experience in the field indicates that the best results are to be obtained from the intra-jugular injection of 5 cc. of coarse grain spleen and gland

pulp, mixed either with aleuronat or peptone, the inoculated animals being exposed to veld infection 14 days later.

C. Inoculation of calves.

The calves used were from 3 weeks to 3 months old.

Experiment 16.—(a) Twenty-two calves were injected with 5 cc. of medium grain pulp mixed with peptone. The calves were obtained from a clean area and they remained there until the 14th day after injection. The percentage of survivors was 31·7.

(b) Eight calves were injected as in (a), but were allowed to graze on infected land immediately after. Five of the calves died, the survivors being 35 per cent.

Conclusion.—The inoculation of calves in the field indicates that the results are not likely to be so good as with full-grown animals.

D. Double injection.

Experiment 17.—Twenty-three animals were inoculated with a fine emulsion, and four months later the 18 survivors were injected with 10 cc. of pulp mixed with peptone. Six of these died as a result of the second injection. The percentage of animals surviving the double injection was 52·1.

Part II.

The immunisation of cattle against East Coast Fever at the Laboratory.

Experiment 18.—Two animals were injected with 5 cc. of fine grain pulp. Both reacted and survived two subsequent infestations with ticks and also natural exposure, without reaction.

Experiment 19.—In this experiment aleuronat was mixed with the fine grain pulp. Eleven animals were used. One showed the presence of plasma bodies and all survived. Six of them were tested with ticks and two died. The four survivors were submitted to natural infection and none died. The other five animals that survived injection were exposed to natural infection and two died of East Coast Fever.

Experiment 20.—In this experiment peptone was mixed with the pulp. Sixteen animals were used and one died. Of the 15 survivors 11 were tested with ticks and 5 died. The surviving six of this batch were exposed to natural infection. One died of redwater.

The four animals that were not tested with ticks were exposed to natural infection and two died, the infection being complicated with redwater in each case.

Experiment 21.—Peptone and aleuronat were together mixed with the pulp in this experiment. Five animals were injected and one died. Of the four survivors one showed plasma bodies. These four animals were exposed to natural infection and three died. Among the 3 that died was the animal that had shown plasma bodies as a result of the injection.

Experiment 22.—The pulp in this experiment was of half-medium grain, and it was injected alone, the dose being 5 cc.

Three animals were used; all survived both the injection and a subsequent exposure to natural infection. In two of the animals plasma bodies were seen after both the injection and the test.

Experiment 23.—Conditions as in Experiment 22 with the addition of peptone. Two animals were used. One died and one did not react. The animal which did not react, and which had been previously inoculated died on subsequent exposure to natural infection.

Experiment 24.—Six animals were injected intravenously with 5 cc. of medium grain pulp. Four died of the disease and one of anaemia. The remaining animal which reacted to the injection proved to be immune when exposed to infection.

Experiment 25.—In this experiment the medium grain pulp was mixed with peptone. Thirteen animals were used. Two died and five showed plasma bodies. When the whole of the survivors were exposed to natural infection four died.

Experiment 26.—The dose of medium grain pulp (5 cc.) in this experiment was added to pulverised gelatin and physiological water. Eight animals were used, and six reacted. One of these died of debility before its immunity could be tested. Two of the survivors died when exposed to natural infection.

Experiment 27.—Three animals were injected with 5 cc. of half-coarse grain pulp. All reacted and survived. They failed to contract the disease when exposed to natural infection.

Experiment 28.—The material used was similar to that in Experiment 27 with the addition of peptone. Fifty-one animals were used. Fourteen died of the disease and three from other causes. Twenty-two reacted showing plasma bodies, seven reacted but without any plasma bodies being observed, and the remaining five failed to react typically. Of the 34 survivors, one died when tested with ticks, five showed plasma bodies and recovered, and the remaining 28 survived the critical period of the test.

Experiment 29.—The following table expresses the results obtained in this experiment.

| No. of animals injected. | Solution of Hydro-Quinine. | Died of East Coast Fever from Injection. | Survived Injection. | Died of East Coast Fever from tests. | Survived critical period of tests. |
|--------------------------|----------------------------|--|---------------------|--------------------------------------|------------------------------------|
| 1 | 0.4 % | 1 | — | — | — |
| 6 | 0.5 % | 3 | 3 | 2 | 1 |
| 10 | 0.6 % | 2 | 8 | 2 | 6 |
| 8 | 0.65 % | 3 | 5 | — | 5 |
| 8 | 0.7 % | 2 | 6 | — | 6 |
| 7 | 0.75 % | 1 | 6 | 3 | 3 |
| 3 | 1 % | — | 3 | 1 | 2 |
| 7 | 2 % | — | 7 | 3 | 4 |
| 3 | 4 % | — | 3 | 3 | — |
| 1 | 5 % | — | 1 | 1 | — |
| 1 | 10 % | — | 1 | 1 | — |
| 4 | 20 % | — | 4 | 4 | — |
| 59 | — | 12 | 47 | 20 | 27 |

From this it is seen that when the strength of the quinine solution exceeded 4 per cent. there was no immunisation, and that the optimum strength appears to be about 0·6 to 0·7 per cent.

Experiment 30.—Eight animals were injected with 5 cc. of half-coarse pulp added to pulverised gelatin and physiological water. All showed plasma bodies and two died. Of the six survivors one died of the disease when exposed to natural infection.

Experiment 31.—Nine animals were injected with 5 cc. of coarse grain spleen pulp. Six died, one reacted and recovered, and the other two did not react. The one which reacted proved to be immune when tested with ticks, but succumbed to natural infection later. Of the two which did not react, one died and the other proved to be immune when exposed to natural infection.

Experiment 32.—In this experiment aleuronat was mixed with the coarse pulp. Eleven animals were injected. Six died, and of the remaining five, two developed the disease but recovered when subsequently tested, and the remainder were immune.

Experiment 33.—Of 5 animals injected with 10 cc. of coarse grain pulp mixed with aleuronat, all survived. Three died of East Coast fever when tested with ticks, and the other two survived tick infestation and exposure to natural infection.

Experiment 34.—Eleven animals injected with 5 cc. of pulp mixed with peptone. Six reacted, and of these 3 died. Two of these were found to be immune to tick infestation, but one contracted the disease naturally and recovered. Of the six animals remaining, one died, one had a reaction, and the rest were found to be immune when exposed to natural infection.

Experiment 35.—Five animals were injected with 10 cc. of pulp mixed with peptone. Two died, and the remainder were found to be immune both to tick infestation and to natural infection.

Experiment 36.—In this experiment the conditions obtaining in Experiment 34 were repeated with the exception that aleuronat was added as well as peptone. Five animals were used. Three reacted to the injection and two died. Of the three survivors two died on exposure to natural infection.

Experiment 37.—In this experiment the animals were given two injections of pulp. In some instances the pulp was given pure, while in others it was mixed with peptone or aleuronat. The intervals elapsing between the injections varied from about three weeks to two and a half months. In one instance the pulp used for the first injection was saturated in a 10 per cent. solution of quinine.

Twelve animals were used. In four cases there was no reaction. In two cases there was a reaction with the discovery of plasma bodies, but the animals recovered. In the remaining cases there was a reaction and the animals recovered. Only one animal died when exposed to natural infection, although several reacted.

Experiment 38.—Twenty-five animals were injected on two occasions with 5 cc. of pulp mixed with peptone and varying strengths of quinine solution. Twelve died of the disease.

Of the 13 survivors, six died when submitted to the immunity test, but as is shown in a table, all these six animals which had not acquired immunity from the injection, had received pulp which had been soaked in hydro-quinine of from 10 to 40 per cent. strength.

Experiment 39.—Twenty-five animals were injected on three occasions with 5 cc. of pulp of varying grains and in varying mixtures. Two died from the injection and 18 proved to be immune when tested.

The following conclusions are drawn:—

"1. The experience in the field indicates that the inoculation can safely be undertaken in respect of either clean or infected cattle with the prospect of conferring immunity on 50-60 per cent.

"2. The best results in the field may be expected by the injection of 5 cc. of spleen and gland pulp (medium, half-coarse, or coarse grain) mixed with Peptone or Aleuronat, such animals to be kept on clean veld for 14 or 15 days before they are exposed to natural infection.

"3. The immunity conferred by the injection may not be absolute, inasmuch as 12 breakdowns were noted among the experimental animals, or 1 per cent.

"4. The animal which supplied the spleen and gland pulp for the injection has apparently an influence on the results, as the variation in mortality from the injection cannot be considered to be due to any other factor.

"5. As a possible improvement to the present method of immunising cattle against East Coast fever, the saturation of the pulp in a solution of Quinine Hydrochloride is suggested, the strength of the solution to be between 0.6 and 0.7 per cent."

TRYPANOSOMIASIS.

(483) BRUCE (D.), HARVEY (D.), HAMERTON (A. E.), & Lady BRUCE. Morphology of Various Strains of the Trypanosome causing Disease in Man in Nyasaland. The Wild-game Strain. —*Proc. Roy. Soc.* 1913. June 12. Series B. Vol. 86. No. B 589. pp. 394-407. With 7 charts.

The trypanosomes used in the investigations described in this paper were isolated by inoculation of blood from a reedbuck, a waterbuck, an oribi, and two hartebeeste. Measurements were taken of 500 trypanosomes in the blood of rats inoculated with the five strains.

The charts obtained closely resembled each other and also those obtained from some of the human strains described in a previous paper.

The following conclusions are drawn:—

"1. The five Wild-game strains resemble each other closely, and all belong to the same species of trypanosome.

"2. The Wild-game strains and the Human strains, although they differ to some extent, also belong to the same species.

"3. This species is *T. rhodesiense* (STEPHENS and FANTHAM).

"4. There is some reason for the belief that *T. rhodesiense* and *T. brucei* (PLIMMER and BRADFORD) are one and the same species."

- (484) BRUCE (D.), HARVEY (D.), HAMERTON (A. E.), & Lady BRUCE. Morphology of various Strains of the Trypanosome causing Disease in Man in Nyasaland. The Wild *Glossina morsitans* Strain. — *Proc. Roy. Soc.* 1913. June 12. Series B. Vol. 86. No. B 589. pp. 408-421. With 7 charts.

The strains from which the charts given in this paper were constructed were obtained from wild flies, and the trypanosomes were isolated by feeding the flies on a number of animals, the trypanosomes being then inoculated into rats, these being solely used for the measurements.

As in the case of the wild-game strains 500 individuals of each strain were measured.

Superposed charts of the Wild-game strain, the Wild-fly strain and the Human strain shew that the last of these differs to a considerable extent from the other two, which closely resemble each other. But it was found that the form of the chart was the only thing which supported the idea that the trypanosomes might not be identical, every other factor indicating identity.

The conclusions are as follows:—

- "1. The five Wild *Glossina morsitans* strains resemble each other closely, and all belong to the same species of trypanosome.
- "2. The Wild *Glossina morsitans* strain, the Human strain, and the Wild-game strain, belong to the same species.
- "3. This species is *T. rhodesiense* (STEPHENS and FANTHAM).
- "4. It is probable that *T. rhodesiense* and *T. brucei* (PLIMMER and BRADFORD) are identical."

- (485) KINGHORN (A.), YORKE (W.), & LLOYD (Ll.). Final Report of the Luangwa Sleeping Sickness Commission of the British South Africa Company, 1911-1912. — *Ann. Trop. Med. & Parasit.* 1913. June 10. Vol. 7. No. 2. pp. 183-302. With 12 plates.

The matter contained in this Report is divided into six sections as follows: The Human Trypanosome; Trypanosomes of Game and Domestic Stock; Trypanosomes in Wild *Glossina morsitans*; Description of the Trypanosomes; Development of *Trypanosoma rhodesiense* in *Glossina morsitans*; and the Report of the Entomologist. Two appendices contain a description of an experiment to ascertain whether Tabanids transmit trypanosomes in nature, and an account of an attempt to transmit *Trypanosoma rhodesiense* by means of *Ornithodoros moubata*.

Much of the matter contained in the report has already been published in the form of separate papers.

The following is the summary of the section dealing with the human trypanosome:

- "1. The human trypanosome (*T. rhodesiense*) is distributed widely throughout South Central Africa.
- "2. There is no essential difference between the clinical manifestations of the disease in man caused by *T. rhodesiense* and that due to *T. gambiense*, except possibly the greater virulence of the former.
- "3. *T. rhodesiense* is transmitted in Rhodesia by *Glossina morsitans*.

"4. Approximately 3·5 per cent. of the flies may become permanently infected and capable of transmitting the virus.

"5. The period which elapses between the infecting feed of the flies and the date on which they become infective varies from eleven to twenty-five days in the Luangwa Valley.

"6. Attempts carried out at laboratory temperature on the Congo-Zambesi plateau during the cold season to transmit the human trypanosome by means of *Glossina morsitans* were invariably unsuccessful in spite of the fact that 680 flies were used in these experiments.

"7. The developmental cycle of *T. rhodesiense* in *Glossina morsitans* is to a marked degree influenced by the temperature to which the flies are subjected. High temperatures (75°–80° F.) favour the development of the parasite, whilst low temperatures (60°–70° F.) are unfavourable.

"8. The first portion of the developmental cycle can proceed at the lower temperatures, but for its completion the higher temperatures are essential.

"9. The parasites may persist in the fly at an incomplete stage of their development for at least sixty days under unfavourable climatic conditions.

"10. These observations afford an adequate explanation of the extremely long latent periods of trypanosomes in *Glossina* which have occasionally been observed by various workers.

"11. The relative humidity of the atmosphere has apparently no influence on the development of the trypanosome in *Glossina morsitans*.

"12. Mechanical transmission does not occur if a period of twenty-four hours has elapsed since the infecting meal.

"13. *Glossina morsitans*, in nature, has been found to transmit the human trypanosome.

"14. The chief reservoir of the human trypanosome is the antelope.

"15. The results of examination for the human trypanosome of the blood of a large number of monkeys, wild rats and mice were invariably negative."

It is estimated that the percentage of big game infected with trypanosomes pathogenic to man and domestic stock in the Luangwa Valley may be placed at 50, and on the Congo-Zambesi watershed at 35.

In the Luangwa Valley six species of trypanosomes were isolated from game and stock, viz., *T. rhodesiense*, *T. vivax*, *T. nanum*, *T. pecorum*, *T. montgomeryi*, and *T. multiforme*. On the Congo-Zambesi watershed the following trypanosomes were isolated: *T. rhodesiense*, *T. vivax*, *T. nanum*, *T. pecorum*, and *T. tragelaphi*.

In the Luangwa Valley *T. rhodesiense*, *T. pecorum*, and a third parasite termed by the authors *T. ignotum* were isolated from wild *Glossina morsitans*. In a foot-note to a subsequent section it is stated that *T. ignotum* is probably identical with the organism described by the Sleeping Sickness Commission of the Royal Society and named *T. simiae*, this name claiming priority should the two prove to be identical.

In the fourth section of the report is given a description of the trypanosomes encountered by the Commission, and it is perhaps convenient to include a brief summary of the section.

Trypanosoma rhodesiense having been dealt with at length in the first section of the report, it is not further described save to mention the varieties of animals from which it was isolated.

T. vivax.—This organism is principally characterised in the living state by its extraordinary rapidity of movement.

In stained preparations it is more or less club-shaped with a long free flagellum. The nucleus is placed about the centre in the body. The blepharoplast is large and rounded, and lies close to the posterior end of the body. The undulating membrane is poorly developed or nearly absent. The average length is 23·6 microns, with a maximum of 28·75 and a minimum of 18·75. The average breadth is 3·2 microns.

In a small number of experiments monkeys, rabbits and rats were found to be insusceptible.

Although it could not be definitely proved that this trypanosome is transmitted by *G. morsitans* in nature, owing to the lack of suitable experimental animals, it was proved experimentally in two cases that the fly is capable of transmitting the parasite.

T. nanum.—A short sluggish organism with no progressive motion.

In stained preparations the posterior extremity of the body is rounded, and the blepharoplast which is small is placed quite close to it. There is no free flagellum and the membrane is poorly developed. The protoplasm is free from granules and vacuoles. The length varies from 19 to 10 microns with an average of 14·3. The average breadth is 1·5 microns. Monkeys, rats and one rabbit were found to be insusceptible.

T. pecorum.—It is stated that owing to the lack of suitable experimental animals this trypanosome could not be distinguished with certainty from the preceding and its morphological characters are practically the same.

While the transmitting agent of *T. nanum* could not be ascertained with certainty, it was proved that *G. morsitans* transmits *T. pecorum* in nature.

T. multiforme, n. sp.—This organism was isolated from a bush-buck at Nawalia. It is remarkably polymorphic and variations of motility are observed, the short sluggish ones showing no tendency to alter their position, while the long forms which are provided with a free flagellum shew marked translatory movement. The maximum length is 33·5 and the minimum 10·5 microns. The organism is said to resemble *T. gambiense* more closely than any other species. Monkeys, rabbits, and rats were found to be susceptible, the guinea-pig proving refractory. The infection in laboratory animals runs a very chronic course. The transmitting agent is unknown.

T. montgomeryi(?).—Found in one dog only on the Nyasaland border. The organism is broad and stumpy, and shows no marked translatory power. The greater breadth of the parasite serves to distinguish it from *T. pecorum*. The blepharoplast lies quite close to the posterior extremity and sometimes projects slightly from the surface. Membrane very poorly developed and there is generally no free flagellum. The posterior portion of the body often contains large vacuoles, and granules may be found scattered through the whole of the cytoplasm. The maximum length is 20 and the minimum 10 microns. The breadth varies from 1·25 to 6·5 microns.

Nothing is known regarding either the pathogenicity or transmission of the organism.

T. ignotum (probably identical with *T. simiae*). A comparatively short slender organism which is fairly actively motile, but with little power of translation.

The posterior extremity is generally rounded and the blepharoplast which is small sometimes projects from the surface. The membrane and flagellum are poorly developed, and there are generally no granules or vacuoles in the body. The maximum length is 23 and the minimum 12 microns.

The organism was isolated from wild *Glossina morsitans*, but it has never been found in wild game. Monkeys were found to be very susceptible, death taking place in from 5 to 16 days. Rats, guinea-pigs, mice, cattle, and dogs failed to become infected. In a goat the trypanosome was once seen, but the animal survived. Rabbits could be infected, but the disease did not as a rule run a rapid course, two surviving over a hundred days.

The percentage of infective flies was found to be not less than 0.3 per cent., and in one instance evidence was obtained that development of the trypanosome occurs in the proboscis.

T. trigelaphi.—Closely resembles *T. ingens*, but appears to be rather shorter and more slender. The possibility is suggested that leeches or mosquitoes may be the transmitting agents.

Section V. of the report deals with the development of *T. rhodesiense* in *Glossina morsitans*.

Section VI. comprises the report of the Entomologist and deals with (a) *Glossina morsitans* in the laboratory, (b) A record of some breeding places of *Glossina morsitans*, (c) A record of blood-sucking insects and ticks collected in the Luangwa Valley and at Ngoa. In collections of *Glossina morsitans* made without discrimination the proportion of females was often found to be as low as 2 per cent., although it was found that taking into consideration a period of twelve months males and females emerged from the pupae in about equal proportions. In the latter half of the year the males slightly predominated, while in the earlier half of the year the reverse was the case. The period of pupation varied from 21 to 88 days, depending to a great extent on the temperature. The shortest periods were observed at a temperature of 86° F. and the longest at 62° F. Very high or very low temperatures were found to have a deleterious effect.

A description illustrated by a number of photographs is given of places in which *Glossina* was found to have bred. In most cases the positions were such that they received sunshine for a number of hours daily. The pupae were nearly always lightly covered with earth.

The following blood-sucking insects and ticks were found: Ixodoidea, *Ornithodoros moubata*. Tabanidae, *T. africanus*, *T. biguttatus*, *T. par*, *T. liventipes*, *T. nigrostriatus*, *T. taeniola*, *T. taeniola* var. *variatus*, *T. fraternus*, *T. fuscipes*, *T. albipalpus*, *T. pullulus*, *T. claritibialis*, *T. atrimanus*, *T. copemani*, *T. diversus*, and *T. maculatissimus*. In addition to these, five other species were collected but have not yet been identified.

Haematopota were very rarely encountered as compared with the foregoing and the following species have been identified: *H. mactans*, *H. insidiatrix*, *H. sp.* (undescribed). Muscidae, Glossina—the only species collected was *G. morsitans*.

Auchmeromyia luteola was commonly found. *Cordylobia anthropophaga* was encountered. A few specimens of *Stomoxys nigra* were found and *Stomoxys calcitrans* was very numerous at times. Pupipara, *Hippobosca hirsuta* was repeatedly found. Two other species of *Hippobosca*, one wingless and the other winged were taken, the former on several occasions, and the latter only once.

In an appendix to the report is given an account of an experiment undertaken to ascertain whether tabanids transmit trypanosomes in nature. The flies were placed, as they were caught, on a monkey, but only a small proportion fed. All that fed were dissected and examined, and only seven out of the total number—128—were found to contain flagellates; these were present in the mid and hind gut only. Inoculations were made into wild rats on four occasions, but in only two of these instances did the rats survive sufficiently long to allow of conclusions being drawn, and these remained negative.

In a second appendix a brief account is given of an experiment carried out with the object of transmitting *T. rhodesiense* by means of *O. moubata*. The experiment failed entirely.

For abstracts of portions of this report published as separate papers see this *Bulletin*, Vol. 1, pp. 78, 79, 95, 149, 155, and 156.

(486) WENYON (C. M.) & HANSCHALL (H. M.). A further Note on *Trypanosoma rhodesiense* from Three Cases of Human Trypanosomiasis.—*Jl. London School Trop. Med.* 1913. April. Vol. 2. No. 2. pp. 123-128.

The observations recorded in this paper were made with particular reference to the occurrence of posterior-nuclear forms, the constancy of their presence in the blood of white rats, their percentage and variation, and the relationship between the virulence and the percentage of posterior-nuclear forms.

The strains were obtained from three cases of infection in the human subject, in none of which were any trypanosomes with the posterior disposition of the nucleus seen. Such trypanosomes were observed in the first rat of each series inoculated from the patients. In each series of rats subinoculated with the strains there were days on which posterior-nuclear forms were absent, and in one rat none were seen during the twelve days that it lived. As a rule the posterior-nuclear forms were observed in the broad trypanosomes, but they were also observed in the long slender forms.

Although the number of observations was too small to allow of definite conclusions being based upon them, the authors got the impression that posterior-nuclear forms were more numerous in the first passages in the rats, and in some cases the proportion of these forms appeared to increase in proportion to the length of the rat's life, but there were numerous exceptions.

No correlation could be established between the number of posterior-nuclear trypanosomes and the virulence of the strains.

In connection with the importance of the posterior-nuclear forms the following paragraphs may be quoted:

"It has been noted that these forms are known to occur in *T. pecaudi* (Soudan) and *T. brucei* (Uganda).

"Recently STEPHENS and BLACKLOCK have expressed the belief that *T. brucei* of Uganda is not the same species as *T. brucei* of Zululand, the original source of the type species of PLIMMER and BRADFORD. They base their belief on the fact that *T. brucei* (Zululand) is a constant monomorphic trypanosome, while *T. brucei* (Uganda) is a markedly polymorphic one—in this resembling *T. pecaudi* and *T. rhodesiense*.

"If this be so, it will probably be found, as some have maintained, that the Uganda *T. brucei* is the same as *T. pecaudi* of the Soudan. The trypanosome of Uganda hitherto known as *T. brucei* would therefore have to be called *T. pecaudi*. Further, there seems to be an accumulating amount of evidence in favour of regarding *T. rhodesiense* as identical with *T. brucei* of Uganda. If this turns out to be correct, then the specific name *rhodesiense* will have to give place to the name *pecaudi*."

- (487) BEVAN (Ll. E. W.). Preliminary Note on a Trypanosome causing Disease in Man and Animals in the Sebungwe District of Southern Rhodesia. — *Il. Trop. Med. & Hyg.* 1913. April 15. Vol. 16. No. 8. pp. 113-117. With 2 plates and 7 temperature charts.

The strains used in the observations recorded in this paper were obtained from a man, a dog, and a goat, two rabbits being inoculated from each strain. From the examination of moist and stained preparations taken from the first passage rabbit in each case it appeared that the trypanosomes were identical. In each case long, short and intermediate form were observed, these gradually merging into each other.

The trypanosomes appeared to correspond very closely with *T. rhodesiense*. The animals inoculated did not show the oedema of the head that characterised the infection produced by the trypanosome obtained from 'W. A.'

A further report is to be published regarding the trypanosomes found in two of the experimental rabbits.

- (488) LAVERAN (A.). Au Sujet du *Trypanosoma rhodesiense* et du *Trypanosoma brucei*.—*Bull. Soc. Path. Exot.* 1913. May. Vol. 6. No. 5. pp. 340-343.

In this paper brief references are made to the various views that have been expressed by different authors regarding the identity of these two trypanosomes and the evidence that has been considered in arriving at the different opinions. Among other points referred to is the question as to whether the *T. brucei*, Uganda, is identical with *T. brucei* (PLIMMER and BRADFORD).

Laveran is of the opinion that further proof must be furnished before it can be considered as certain that *T. rhodesiense* and *T. brucei* (Uganda) are identical, and suggests that cross-immunity experiments would furnish the most valuable evidence.

27 MAY 1925
M. I. P. U. S.

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ERRATA.

Vol. 13, No. 1, p. 1. Van der Elst Abstract, last line, and in title of Walraven's paper, *for Trypanosoma rodhain read Trypanosoma rodhaini*.

Vol. 13, No. 1, p. 6, 14th line from bottom, name of first author of paper on *Eimeria utinensis* for Lelau (U.), *read* Selan (U.).

Vol. 13, No. 1, p. 13. Thornton Abstract, line 5, *for* Phaecocaeus *read* Phacochoerus.

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- (489) SHILSTON (A. W.). Notes on Zululand Trypanosomes.—*Second Report of the Director of Veterinary Research, Union of South Africa. October, 1912.* pp. 345-361. With 2 plates. (1913. Cape Town: Cape Times, Ltd., Government Printers.)

After discussing at some length the question of the identity of the different strains of trypanosomes known as *T. brucei*, the author proceeds to the description of a trypanosome isolated from a mule in Zululand.

Towards the end of 1911 blood smears made from a beast and from a donkey were found to contain small trypanosomes, but the infected animals died before it was possible to make sub-inoculations. Later, in the hope of obtaining this trypanosome a dog was inoculated with the blood of a sick mule. This dog shewed trypanosomes in its blood six days after inoculation. On making an examination of smears from this animal it was found that it was not the small trypanosome that was present, but an organism in which a considerable proportion of individuals were furnished with a well-marked free flagellum. It seemed probable that the disease was nagana. Further examination shewed that it resembled far more closely the strain known as the Uganda *T. brucei*. A number of charts drawn up according to Bruce's plan are given, and these shew very marked differences, but no great importance can be attached to them as they have not been constructed upon the examination of a very large number of organisms, nor have they been made methodically with regard to the period of infection, etc.

The author draws the following conclusions:—

1. That a trypanosome occurs in animals in Zululand, which is markedly dimorphic, and in this and other respects closely resembles the trypanosome described by Sir David Bruce and others as *Trypanosoma brucei*.
2. That the trypanosome introduced into Europe in 1896 and described by various authorities as *Trypanosoma brucei*, differs to a marked degree from that described under the same name by Bruce, and from the Zululand trypanosome described in the present paper.
3. That a careful study of the trypanosomiasis of Zululand is necessary to determine, among other things, whether a trypanosome having the characters of *T. brucei* as formerly described, also exists in that country.

Notes on a small trypanosome from Zululand.

The small trypanosome referred to in the preceding note has been frequently observed in blood smears from widely different sources, but it has not been possible as yet to obtain a strain for accurate study. The organism has been observed in the blood of oxen and donkeys.

In preparations fixed in alcohol and stained with Giemsa the maximum length is 13 microns and the minimum 9. The breadth varies from 1.5 to 2.5 microns. The body of the parasite is short and stout. The posterior extremity is rounded and the anterior tapering. A few chromatin granules may be present in the anterior portion of the body, but frequently the cytoplasm stains uniformly. The nucleus is of a rather elongated oval shape, and situated about the centre of the body. The micronucleus is round and placed close to the posterior extremity. The undulating membrane is not well developed and there is no free flagellum.

Since no animal inoculations have been carried out as yet, it is impossible to make any definite pronouncement as to the identity of the organism, but it appears to resemble *T. pecorum* very closely. The fact that it has been found in donkeys excludes *T. nanum*, and *T. uniforme* would appear to be excluded by its greater average length, 16 microns.

- (490) RONDONI (P.) & GORETTI (G.). *Richerche sperimentali sul Nagana. I. Comunicazione. Su alcune Proprietà Biologiche della Milza nella Infezione sperimentale da Trypanosoma brucei.* [Experimental Researches on Nagana.]—*Lo Sperimentale*. 1913. April 7. Vol. 67. No. 1. pp. 1-24.

This paper contains a detailed account of experiments which were carried out by the authors to ascertain whether trypanocidal substances and specific antibodies are formed in the spleen during infection, and also to investigate the haemolytic powers of spleen extract.

The general conclusions arrived at are:—

The spleen of animals (guinea-pigs and rats) infected with *T. brucei* sometimes contains trypanolytic substances; this organ appearing to play a special part in the defence of the organism.

Aqueous extracts of the spleen of infected animals were found to possess a haemolytic power for red corpuscles obtained from any source, and even from the same individual. The substances can be extracted with alcohol and they behave in different manners when subjected to heat. Normal serum appears, at least in part, to exert an inhibiting effect. They are probably of a lipid nature and seem to be of very little vital importance.

- (491) MARTOGGIO (F.). *Sulle Tripanosomiasi del Dromedario Eritreo.* [Trypanosomiasis of the Dromedary in Eritrea.]—*Annali d'Igiene Sperimentale*. 1913. Vol. 23. No. 2. pp. 229-234. With 2 text-figures.

There are two trypanosomes to which the dromedary is susceptible. One of these is also the cause of a disease in bovines, which is termed "giahān" and the other which is common to the ox, horse and sheep is responsible for the disease known as "atfeh," and belongs to the surra group of trypanosomes.

The trypanosomes are distinguishable by the following characters:—"Giahān" is transmitted by Stomoxys, while "atfeh" is transmitted by Tabanidae. The two organisms are morphologically distinguishable. That causing "giahān" appears to be dimorphic, as it is said that young forms measure from 14 to 16 microns and adult forms 24 microns including the flagellum. The parasite of "atfeh" measures about 30 microns excluding the flagellum, which measures about 8 microns. The parasite of "giahān" is not pathogenic for the dog, rabbit, mouse, whilst that causing "atfeh" is pathogenic for these animals. The parasite of "atfeh" has little or no pathogenic power for the adult ox and the sheep, while that of "giahān" is markedly pathogenic for these animals.

- (492) KOHL-YAKIMOFF (Nina), YAKIMOFF (W. L.), & BEKENSKY.
 Le Trypanosome des Bovidés (*Tr. theileri* ou du Type voisin)
 en Russie d'Europe. [The Trypanosome of Cattle (*Theileri*
 or a similar Type) in Russia in Europe.]—*Bull. Soc. Path.*
Exot. 1913. June. Vol. 6. No. 6. pp. 433-434.

The authors have examined the blood of 14 animals without finding flagellates by microscopic examination, but by employing SERGENT's method have been able to obtain cultures of flagellates from 5 of them. They have been able to pass the strain on from the broth to NNN medium and keep it for several generations. They have also been able to inoculate a calf successfully.

- (493) KOHL-YAKIMOFF (Nina), YAKIMOFF (W. L.), & SCHOKHOR
 (N. J.).—Le Trypanosome des Bovidés (*Tr. theileri* ou du Type
 Voisin) au Turkestan. [Trypanosome (of the *Theileri* or
 allied Type) in cattle in Turkestan.]—*Bull. Soc. Path. Exot.*
 1913. June. Vol. 6. No. 6. p. 434.

The authors have obtained cultures of the trypanosome from the blood of cattle in eight cases out of nine examined. The cattle in Turkestan are said to be very frequently infected with *Theileria* (*Piroplasma*) *mutans*.

- (494) FRY (W. B.) & RANKEN (H. S.). Further Researches on the
 Extrusion of Granules by Trypanosomes and on their further
 Development. (With a Note on Methods by H. G. PLIMMER.)
 —*Proc. Roy. Soc.* 1913. June 12. Series B. Vol. 86.
 No. B. 589. pp. 377-393. With 3 plates.

Two methods were principally used in the earlier part of the investigations described in this paper. These were dark-ground illumination and vital staining with a 0.75 per cent. solution of toluidine blue in physiological salt solution. Other methods have been devised and these are described at some length.

Two types of granules are described as occurring in the bodies of trypanosomes, namely, reserve food granules and the special granules which are the subject of this communication. The latter granules are of nuclear origin and are infective.

These infective granules have been found in *T. gambiense*, *T. rhodesiense*, *T. brucei*, *T. evansi*, *T. nanum*, *T. pecaui*, and *T. lewisi*.

When examined with dark-ground illumination the granule appears as a small, sharply-defined, highly refractile body, and with vital staining it takes up the stain rapidly and acquires an intense colour.

The number of granules varies in different species of trypanosomes, and the number and size of granules present in any species are inconstant. It was observed in the case of *T. nanum* carried on in gerbils, that the granules increased in number but decreased in size as the virulence of the trypanosome increased.

The authors have found that granules are not always necessarily present in trypanosomes, nor have they been able to determine what conditions are necessary for their formation, but they are able to indicate at what stage they make their appearance.

It was observed that when trypanosomes first made their appearance in the blood they did not contain granules. After about the fourth day granules were to be observed in the trypanosome bodies and for about 24 hours trypanosomes containing granules were numerous. After this period when free granules were numerous in the blood the proportion of trypanosomes containing granules decreased until, finally, although the trypanosomes might be enormously numerous in the blood, none of them contained granules.

This was found to hold good for *T. brucei*, *T. nanum*, and *T. evansi*.

The original observations regarding the extrusion of granules have been confirmed and the process has been followed with *T. nanum* and *T. gambiense*. The former is very suitable for the purpose as the granules are large and the trypanosome does not execute any marked translatory movements.

When extrusion of a granule is about to take place it passes from the centre of the body towards the pointed end and then makes its way back again to the centre. This may occur a number of times, the granule being finally extruded from the pointed extremity.

In *T. gambiense* the granules are multiple and extrusion does not occur at the pointed extremity but from a point near the middle of the body.

Extrusion is most readily observed in infections running an acute course.

The extrusion of the granules may be stimulated by drugs, and when trypanosomes are killed by trypanocidal agents the granules are not necessarily killed but make their escape from the degenerating bodies.

There appear to be three methods by which granules may make their escape. They may be forcibly extruded by the activity of the trypanosome; by active movement of the granule in a rapidly degenerating trypanosome; and by outside agencies such as currents breaking up degenerating trypanosomes and allowing granules which cannot escape by their own movements to be liberated.

Changes of osmotic pressure also have effects upon the liberation of the granules.

With vital staining free granules take the stain rapidly and uniformly. For a short time the freed granule remains stationary near the parent trypanosome, but then shows independent movement, passing across the field with a rolling motion. The authors believe that a pseudopodial process makes its appearance.

The free granules are to be found in the blood, glands, and internal organs. They have been found in the proximal glands in experimental animals 24 hours after infection.

In a foot-note by PLIMMER it is stated that the motility of the granules can be demonstrated by the addition of a little cherry-gum to a preparation as this stops all Brownian movement while it only slows the true motility, and their affinity for toluidine blue assists in distinguishing them from other particles such as blood platelets.

The subsequent stages of development of the granules are more difficult to follow as they cannot be seen in any individual preparation, and consequently the authors are not in a position to state definitely that they have arranged the changes observed in the correct chronological order.

Granules have been observed to have been ingested by leucocytes, and older granules have been observed in the interior of endothelial and other cells, but in these cases they did not appear to have undergone any degeneration.

The free granules increase in size, become oval, and their protoplasm becomes differentiated into a deeply staining central piece and a peripheral portion which stains more faintly. There is also developed a flagellum-like projection. Later the chromatin portion divides unequally.

From this point development appears to proceed in two different directions. In some of the parasites the body becomes elongated, the two masses of chromatin become separated from each other, and the true flagellum is developed. There is no undulating membrane.

In other parasites the body remains rounded in shape and the chromatin masses undergo division, each pair of large and small masses becoming provided with a flagellum attached to the smaller and projecting beyond the cytoplasm.

These forms have been found in man and in animals in preparations from glands, internal organs, and bone-marrow.

Attempts were made to infect animals with materials containing granules only. Blood containing large numbers of trypanosomes and granules was mixed with two volumes of 2 per cent. salt and 1 per cent. citrate solution and left at temperatures of from 34° to 38° C. for an hour. At the end of that time no trypanosomes could be found intact, they had all become swollen and rounded, and the still active granules had escaped.

It was found possible to infect gerbils in this way with *T. nanum*, trypanosomes appearing in the circulation in about 5 days.

In some instances the gerbils were killed before a trypanosome appeared in the circulation with the object of tracing the stages of development of granules into trypanosomes. In these animals granules and later stages were found in proximal glands and internal organs.

It was found to be impossible to stain the developmental stages of the granules by ordinary methods, but success was obtained with special methods devised by PLIMMER.

As an addendum to the paper there is a note by PLIMMER on a "New method of Blood Fixation."

There are two methods recommended, namely, fixation with the vapour of a solution of iodine in chloroform, and fixation with salt solution containing potassium iodide and iodine to saturation. It is said that there is much less distortion, and that there is finer fixation of details by these methods. The caryosome in the nucleus, the distinction between the micronucleus and blepharoplast, and the details of dividing forms are said to be far more clearly made out.

Vapour method.

The thinnest possible films are exposed to the vapour of iodine in chloroform for 10 to 15 seconds while still moist. When dry they are placed in chloroform or a mixture of ether and alcohol in equal parts for two hours. The preparations may then be stained in Giemsa, made by mixing one drop of Giemsa solution with two of distilled water, for 2 to 12 hours. They are then washed and treated with orange tannin solution for 15 seconds, again washed thoroughly, dried, and mounted in either cedar oil or liquid paraffin.

They may also be stained with carbol fuchsin for 2 to 12 hours, washed, treated with alcohol until the bulk of the stain is removed, differentiated with clove oil saturated with orange G, washed with xylol and mounted as before.

Fixation with iodine solution.

A saturated solution of iodine and potassium iodide in salt solution is made and 5 or 6 drops of this are added to 10 cc. of salt solution. This is mixed with the blood in equal parts and thickish films made. When the films have begun to dry they are placed in alcohol and ether for two hours and the succeeding steps are the same as those described in the vapour fixation method.

- (495) KLEINE (F. K.) & ECKARD (B.). Ueber die Bedeutung der Speicheldrüseninfektion bei der Schlafkrankheitsfliege (*Glossina palpalis*). [The Significance of the Infection of the Salivary Glands in *Glossina palpalis*.] — *Zeitschr. f. Hygiene u. Infektionskrankh.* 1913. April 25. Vol. 74. No. 1. pp. 183-187.

In this paper are recorded the results of experiments in which a number of apes were inoculated with the contents of different portions of flies infected with *T. gambiense*.

Six apes were inoculated with the contents of the salivary glands and all these became infected. A second series of six were inoculated with the intestinal contents of the same six flies and in no case did trypanosomes appear in the blood. Three apes were inoculated with the contents of the proventriculus but these also failed to become infected. It would therefore appear that the infection of the salivary glands is an essential stage of the development of the trypanosome in the fly, and not in the nature of an accident.

- (496) ROUBAUD (E.). Evolution comparée des Trypanosomes pathogènes chez les Glossines. [The Comparative Development of Trypanosomes in Glossinae.]—*Bull. Soc. Path. Exot.* 1913. June. Vol. 6. No. 6. pp. 435-441. With 3 text-figures.

In this paper an attempt is made to devise a classification of the African trypanosomes based upon the manner in which the different species undergo development or maintain their existence in the flies.

According to the author there are two main classes, viz., those in which the trypanosomes are merely "cultivated," and those in which the trypanosomes undergo a process of "evolution" in

the fly. In the former the characteristic feature is the persistence of the trypanosome form, and in the latter there is a transformation from the trypanosome form into the crithidial type, which again changes into the typical trypanosome. Either of these conditions may be temporary or lasting.

In "temporary cultivation" the trypanosomes pass into the posterior portion of the midgut where the cultivation occurs, giving rise to no important modifications of morphology; there being as a rule an increase in size only. The process generally lasts only two or three days and occurs in the ingested blood only, disappearing when this disappears. The author states that this method of development is constant for the following viruses: *T. gambiense*, *T. dimorphon*, *T. congolense*, and *T. pecaudi*.

He has never observed it with the following viruses: *T. congolense*, *T. brucei* (Zululand strain at the Pasteur Institute), *T. cazalboui*, and *T. evansi*. [There appears to be an error in this statement as *T. congolense* occurs in both groups.]

This "intestinal cultivation" may in some cases become lasting, and may persist in the intestine even during a period of fasting, the parasite extending forwards to the anterior portion of the gut. There is never any infection of the rectum or of the Malpighian tubes, and cysts are never formed. When the whole of the mid- and fore-gut is invaded the author terms the invasion "infection totale."

While "temporary intestinal cultivation" is constant for the species mentioned, "lasting intestinal cultivation" occurs in a small proportion of flies only. This depends upon the species of *Glossina*, the locality, and the specific character of the virus. In the case of *T. dimorphon* in *G. palpalis* in Dahomey it was found in 1 per cent. of flies only.

This lasting infection of the intestinal tract must be considered as a "cultivation" because the morphological modifications which occur are very slight and closely resemble those seen in artificial cultures.

Finally, these forms are not inoculable to the vertebrate host.

"Evolution" in the fly is marked by the transformation of the parasites into crithidial bodies which occur solely in the salivary apparatus. This process may also be temporary or lasting.

The "lasting salivary evolution" may be divided into three types:—(1) That described as "direct fixation in the proboscis." In this type of evolution the parasites attach themselves directly to the wall of the proboscis in the form of short crithidia which develop into the salivary trypanosomes, these passing to the hypopharynx. (2) That described as "indirect fixation in the proboscis." In this type of evolution the infection of the proboscis is not derived direct from the blood ingested, but results from extension forwards from an "intestinal cultivation." This type of infection has been observed by the author in the case of *T. dimorphon* and *T. pecaudi*. These two trypanosomes are distinguishable by the forms taken in the hypopharynx. In the case of *T. dimorphon* they are short and devoid of a free flagellum, and in the case of *T. pecaudi* they are long and are provided with a flagellum. (3) Evolution by "indirect fixation in the salivary glands." This method of development

is peculiar to the two human trypanosomes. Infection of the proboscis in this instance also results from "*infection totale*," but the forms which invade the proboscis pass on to the salivary glands and there develop into typical trypanosomes.

From the biological point of view it is interesting to note that development by "direct fixation" in the proboscis only corresponds with a shortening of the cycle of passage in the fly and tends towards mechanical transmission. *T. cazalboui*, the evolution of which is the most simple, appears to be most easily transmitted mechanically.

"Lasting evolution" of *T. cazalboui* in the proboscis only has been observed by BOUFFARD in *G. palpalis* and *G. tachinoides*, and by BOUET and ROUBAUD in *G. morsitans* and *G. longipalpis* in addition, and by the members of the Belgian mission to Katanga in *G. morsitans*.

BRUCE and his collaborators have observed it in the case of *T. vivax* in *G. palpalis*, and *T. caprae* in *G. morsitans*, and it has been observed by FRASER and DUKE for *T. uniforme*. The author thinks that the absolute similarity of the method of development of these trypanosomes strongly suggests their "specific unity."

The author believes that four fundamental types of trypanosomes can be distinguished, basing the distinction on the evidence afforded by the method of multiplication in the fly.

These four groups are:—

The *cazalboui-vivax* type.

The *dimorphon-pecorum* type.

The *pecaudi* type.

The *gambiense-rhodesiense* type.

(497) BRUCE (D.), HARVEY (D.), HAMERTON (A. E.), & Lady BRUCE. Infectivity of *Glossina morsitans* in Nyasaland.—*Proc. Roy. Soc.* 1913. June 12. Series B. Vol. 86. No. B 589. pp. 422-426.

In the experiments recorded in this paper 10,081 flies in batches were fed each batch upon three healthy animals in succession on three consecutive days, and the feedings were repeated three times. The average number of flies in each batch was about 60, so that each animal might possibly be bitten by 180 flies.

In a table are given the results of these feeding experiments upon monkeys, dogs, and goats, and the species of trypanosomes found. The results showed that the following four species of trypanosomes are carried by *G. morsitans* in the district:—*T. brucei vel rhodesiense*, *T. pecorum*, *T. simiae*, and *T. caprae*.

T. brucei vel rhodesiense was found in 2 per 1,000. *T. pecorum* in 4.6 per 1,000, *T. simiae* in 3.4 per 1,000, and *T. caprae* in 3.5 per 1,000.

Infective flies occur at all seasons in about the same numbers.

The following are the conclusions drawn from the experiments:—

"1. The tsetse flies (*Glossina morsitans*) caught in the 'fly-country' near Kasu are infected with four species of disease-producing trypanosomes—*T. brucei vel rhodesiense*, *T. pecorum*, *T. simiae*, and *T. caprae*.

" 2. The proportion of infective flies is 13.5 per 1,000.

" 3. The proportion of flies infective with *T. brucei* vel *rhodesiense*, the cause of the Human Trypanosome Disease in Nyasaland, is 2 per 1,000.

" 4. The flies are found infective all the year round.

" 5. To prevent the infection of tsetse flies it is proposed that the experiment should be tried of destroying all the wild game in the "Proclaimed Area" of Nyasaland."

(498) MITZMAIN (M. B.). *The Mechanical Transmission of Surra by Tabanus striatus*, Fabricius.—*Philippine Jl. Sci. Sec. B. Trop. Med.* 1913. June. Vol. 8. No. 3. pp. 223-229.

The author's success in transmitting surra by means of *Tabanus striatus* has already been referred to in this *Bulletin* (see abstract of MS. letter, Abstr. No. 275, Vol. 1, No. 3, May, 1913).

The majority of the flies used in the experiments recorded in this paper were laboratory bred, but in some instances the flies were obtained from larvae taken from their natural habitats, and in a few instances captured adult flies were used.

Experiments on the direct transmission of the trypanosome were alone carried out. The flies were first allowed to bite an infected guinea-pig or horse for not more than one minute, and were then transferred within five seconds to three minutes to a healthy animal to continue the feed.

A table shows that sixteen experiments were carried out. In these, one and two, and in one instance six flies were used. They were allowed to bite an infected animal as stated, and were then transferred to either a monkey, guinea-pig or horse. In three cases a positive result was obtained. One monkey became infected after it had been fed upon by three flies, and trypanosomes appeared on the eighth day. Its blood infected two guinea-pigs and a horse. In the second successful case a horse was infected by the bites of two flies. A mule, two guinea-pigs and two monkeys reacted to inoculation with this animal's blood. In the third successful case six flies were used to infect a horse. Two monkeys and two guinea-pigs were infected from this animal.

An attempt was made to get Tabanidae to bite animals confined in a large cage. During a period of about six weeks 2,087 female tabanids were liberated in the cage which contained two infected carabaos and one healthy one. The flies died within a few days when kept in the enclosure, and the result of the experiment was negative.

In order to exclude the possibility of hereditary transmission an experiment was carried out with 74 flies derived from eggs laid by a fly which had twice fed upon a heavily infected monkey. The result was entirely negative.

In an experiment with five guinea-pigs an attempt was made to transmit the infection by means other than biting. It was observed that flies placed on an abraded surface would take in surface moisture before biting, and that a fly could obtain sufficient material to fill the stomach in this way without biting.

Areas of skin on healthy and infected guinea-pigs were abraded with a razor and two or three flies were allowed to suck

blood from the surface for about a minute and were then transferred to the healthy guinea-pigs to finish their meal. In no instance was infection obtained.

When flies which had had a meal on an infected animal were used for the preparation of suspensions which were inoculated into guinea-pigs positive results were obtained when the interval elapsing between the infecting feed and the preparation of the emulsion did not exceed 10 hours.

The author's conclusions are as follows:—

"1. *Tabanus striatus* Fabricius for the first time recorded has been found to play a rôle in the transmission of surra. Bred horseflies have been employed for the first time in such experiments. Errors resulting from naturally infected wild flies have thus been eliminated.

"2. Three experiments were successful in the direct or mechanical transmission by 'interrupted' feeding when only a short interval was allowed between the bites on infected and healthy animals. In 16 experiments the minimum number of flies with which the infection could be transmitted was 2.

"3. Trypanosomes of surra were not found to be transmitted hereditarily by *Tabanus striatus* Fabricius.

"4. The contaminated labellum of the fly does not appear to be a factor in the conveyance of the infection.

"5. The maximum length of time that *Trypanosoma evansi* has been demonstrated microscopically in the gut of this species of fly after feeding on infected blood is thirty hours; the organisms were found in the fly's dejecta two and a half hours after biting the infected animal; and suspensions of flies, when injected subcutaneously, were found infective for animals for a period of ten hours after the flies had fed on infected blood."

(499) WENYON (C. M.). Experiments on the Transmission of *Trypanosoma lewisi* by means of Fleas.—*Jl. London School Trop. Med.* 1913. April. Vol. 2. No. 2. pp. 119-123.

In a previous paper (see this *Bulletin*, Vol. 1, No. 3, p. 171) the author recorded the results of his experiments in which it was found that *T. lewisi* is capable of developing both in the human flea (*Pulex irritans*) and in the dog flea (*Ctenocephalus canis*). In the present paper an account is given of further experiments with the human flea, in which it was found that the rat is apparently not a suitable host for the human flea as it does not willingly feed, and soon disappears. It therefore seems improbable that *Pulex irritans* plays any part in the transmission of *T. lewisi* in nature, although it has been shown that its faeces are infective when it has been fed upon an infected rat.

In a further experiment it was proved that *Xenopsylla cheopis*, the common rat flea in India, is a true host of *T. lewisi*; a strain of the trypanosome being carried through three generations in rats by means of three fleas of this species. The periods elapsing before infection took place varied from 9 to 25 days, indicating that the infection is due to some chance such as the licking up by the rat of moist flea faeces. In cases in which the infective faeces were taken direct from the flea and introduced into the mouth of a rat the period of incubation was six to seven days.

The author disagrees with the view expressed by MINCHIN and THOMSON that infection takes place through the agency of small trypanosomes which have wandered from the rectum to the mid-gut and are regurgitated into the rat. His reasons are as follows:—He has not obtained positive results in any experiments in which the possibility of faecal infection has been excluded. The presence of a few small trypanosomes in the mid-gut is to be explained by the very vigorous peristaltic action of the intestine. Another point is that the final development of the infective forms takes place in the rectum rather than in the anterior portion of the gut, thus suggesting that it is the material from the hind-gut which is the source of infection.

- (500) HECKENROTH (F.) & BLANCHARD (M.). *Transmission du Trypanosoma gambiense par des Moustiques (Mansonia uniformis)*. [The Transmission of *T. gambiense* by Mosquitos (*Mansonia uniformis*).]—*Bull. Soc. Path. Exot.* 1913. June. Vol. 6. No. 6. pp. 442-443.

It was recorded in 1908 by MARTIN, LIEBOEUF, and ROUBAUD that they had been able to transmit *T. brucei* from one cat to another by means of *Mansonia*, and by FÜLLEBORN and MAYER in the previous year that trypanosomes (species not stated) could be transmitted from one animal to another by *Stegomyia* by interrupting a feed upon an infected animal and placing the mosquitos in contact with a healthy animal to finish their feed.

The two experiments recorded in the present brief paper were carried out with a strain of *T. gambiense* which had become adapted to multiplication in small rodents.

In the first experiment a healthy and an infected guinea-pig were placed in a cage divided into two parts by a partition reaching to two-thirds of the height. Sixty-four wild *Mansonia* were placed in the cage and the infected guinea-pig was removed at the end of forty-eight hours. Three *Mansonia* were found to be alive at the end of the experiment and these were dissected without any flagellates being found. The healthy guinea-pig was found to be infected on the 22nd day.

In the second experiment an infected guinea-pig was placed in the cage with 26 *Mansonia*. After 24 hours the guinea-pig was withdrawn and after a further interval of 24 hours a healthy guinea-pig was placed in the cage. The guinea-pig was left in the cage for 23 hours and then withdrawn. The animal was found to be infected on the 18th day. No trypanosomes were found in the 6 surviving mosquitos.

- (501) LEESE (A. S.). *Some more Successful Experiments on the Treatment of Surra in the Camel with recommendations for Systematic Treatment*.—*Memoirs of the Department of Agriculture in India. Veterinary Series.* 1913. April. Vol. 1. No. 3. pp. 149-176.

This paper deals with further trials made with certain methods of treatment the details of which have already been published (see this *Bulletin*, 1912, Oct, Vol. 1, No. 1, Abstract 9, pp. 19-21), and with modifications of these treatments. Detailed

descriptions are given of the modifications employed and the results obtained are given and the paper closes with a revised set of rules for the treatment of surra in camels.

Leese after extensive experimentation advises the following method of treatment for camels, and gives the following as an "average" treatment (Treatment "668") :—

| | |
|--------------------------------------|------------|
| 1st day, Soamin subcutaneously | ... 5 g. |
| 2nd day, Tartar Emetic intravenously | 0.5 g. |
| 3rd day, Arsenious Acid | ... 0.6 g. |

After an interval of two days this routine is repeated, increasing the dose of soamin to 6 grammes.

After further 4 days' interval it is again repeated with 6.5 g. soamin.

After a third interval of 8 days repeat with 6.5 to 7.5 g. soamin, the other days in constant quantities throughout. This method is recommended for camels that are in fairly good condition, and in the experiments it yielded 62 per cent. of cures, 22 per cent. of accidents and 16 per cent. of relapses.

Special emphasis is laid upon the fact that the dosage must be carefully graduated to size and weight of the animal. The quantities given are suitable for a camel weighing from 1,050 to 1,300 lbs. Weak and emaciated camels in which the disease has progressed too far to allow them to stand the above treatment should be subjected to the following, which is practically a matter of "kill or cure" (Treatment "178"). It has yielded cures, accidents, and relapses in about equal proportions. As originally described the method was as follows :—

1st day, 55 grains of sodium arsenate per os.

2nd day, 5 grammes of atoxyl subcutaneously.

3rd day, 1.5 g. antimony tartrate intravenously.

Repeated after 3 days' interval.

The conclusions arrived at are as follows :—

1. The best methods of treatment are :—

(a) "668" treatment which gave 62 per cent. of cures, 22 per cent. of accidents, and 16 per cent. of relapses.

[The "668" method is a modification of the "436" method with the sodium arsenate per os replaced by arsenic intravenously.]

(b) "264" treatment which has given over 50 per cent. of cures when applied to strong camels. It is unfit for weak ones, but does not appear to be dangerous to the strong.

(c) "178" treatment which is very dangerous, but is the only hope for the camels which are too weak to stand a longer and better treatment. It cured 35 per cent. and killed 30 per cent.; the remainder relapsed.

2. Soamin in the place of Atoxyl and in exactly the same doses gave identical results, and as it is much cheaper it seems the best of the two drugs to use.

3. Arsenic given intravenously in solution in the place of sodium arsenate given by the mouth, acts as well or better in the longer treatments as regards curative action, while it is less violent in its action on the camel.

4. If a first treatment fails, a second may be applied provided the camel is in a condition to stand it, in which case the chances of cure do not seem to be reduced; from the above account it will be seen that a number of camels have been cured at the second attempt.

5. I have confidence now in saying that a camel is cured on the evidence of 6 months' daily blood-examination since the last dose of the treatment

whenever it is obvious either from blood-examination for a time prior to treatment or by the history of the camel, that the disease was in its acute or sub-acute stage before treatment began. The longest-delayed relapses have been 88 and 89 days, and I have had about 30 camels which have been kept for varying periods under daily blood-examination after that length of time, between 6 and 17 months since treatment and none have ever relapsed. All the first 9 camels cured were, moreover, tested by inoculation of their blood into guinea-pigs.

- (502) MOUCHET (R.) & DUBOIS (A.). *Note sur le Traitement des Trypanosomiasés Animales.*—*Bull. Soc. Path. Exot.* 1913. July. Vol. 6. No. 7. pp. 533-539.

The following is a summary of the authors' conclusions:—

Trypanosoma congolense is very resistant to drugs. The authors have not been able to test the trypanosan which gave good results with RODHAIN, PONS, BEQUAERT, and VANDENBRANDEN. Trypasafrol is useless for the treatment of cattle and no conclusions have been arrived at in the case of goats treated with large doses.

Orpiment appears to have some effect, but it is dangerous.

Trypanosoma cazalboui is resistant to arsenic, but is susceptible to antimony. Trypasafrol has been used in very small doses but without success.

Emetic may produce apparent recoveries lasting for more than two months. It is advised that arsenic be administered at the same time in order to facilitate flattening and maintain strength. Five milligrammes per kilogramme may be injected intravenously with safety, and 7.5 mg. subcutaneously.

Yvon's emetic may be used in slightly larger doses, but it cannot be said that it has any advantages over ordinary emetic.

- (503) ANDREWS (W. H.). *Some Experiments on the Drug Treatment of Trypanosomiasis.*—*Second Report of the Director of Veterinary Research. Union of South Africa. October, 1912.* pp. 362-383. (1913. Cape Town: Cape Times, Ltd., Government Printers.)

The experiments detailed in this paper were carried out with a number of strains of trypanosomes of the *congolense* (or *pecorum*) type. The various strains used were found to possess about an equal pathogenic power. In the earlier experiments horses and mules were largely used, but subsequently sheep were for the most part employed.

In horses the parasite generally appeared in the blood about the 21st day. Only two experiments were made with cattle, and the trypanosome appeared to be rather more pathogenic for them than for horses. In these the trypanosomes appeared in the blood on the 11th day and the animals died on the 43rd and 95th days. In sheep the average period elapsing before the appearance of trypanosomes in the blood was about six days. The duration of the disease varied greatly. About 50 per cent. of the sheep died within a month, while about 10 per cent. survived for a year. In dogs the trypanosomes made their appearance in the blood in from 9 to 15 days, and the animals died in from 15 to 36 days after inoculation.

It was recognised that the desired results could not be obtained from a single trypanocidal agent and a combination of two or more drugs was therefore used. There were made, however, a number of observations on the effects obtained with certain agents used alone.

Arsenophenylglycin.—Nine infected horses and mules were put under treatment, and in every case there was a relapse as indicated by subinoculations. The dose varied from 0.57 to 0.06 g. per kilogramme body weight. In view of the fact that the maximal therapeutic dose of the drug as fixed by EHRLICH is 0.05 g. it is evident that the dose could not be increased with safety.

The two cattle were given 0.059 and 0.074 g. per kilogramme body weight, but in neither case was the treatment effective.

Fourteen sheep were treated. In eleven of these which had received doses of 0.057 to 0.075 g. per kilogramme there was a relapse, generally about the third week. One sheep was given 0.06 g. per kilogramme, and there was no return of trypanosomes up to the 15th day, when further treatment was instituted.

Two sheep which were given 0.09 and 0.0617 g. per kilogramme were apparently cured. The results did not appear to be at all affected by the interval elapsing between the appearance of trypanosomes in the blood and the administration of the drug.

Novoflavin.—Six equines received a dose of 0.01 g. per kilogramme, and one mule was given 0.0098 g. per kilogramme. In three instances the animals treated were suffering from relapses after treatment with arsenophenylglycin, and in these cases there was no return of trypanosomes to the circulation, but the animals were killed about two months after the administration of novoflavin on account of debility.

One of the animals which had not been previously treated was found to be infected by subinoculation on the 169th day, and the remaining two were killed on the 11th and 25th days after treatment owing to debility. No relapse had been observed.

Eleven sheep were given doses varying from 0.009 to 0.013 gramme per kilogramme. In three cases the treatment proved fatal; two animals died shortly after from pneumonia, and in the remaining animals which had been unsuccessfully treated with arsenophenylglycin relapses occurred. It would appear that 0.01 g. per kilogramme is about the maximal dose for both horses and sheep as some fatal results followed doses of this magnitude in both species.

Salvarsan or "606."—Fifteen infected sheep were treated. In the first three cases the animals received salvarsan after unsuccessful treatment with arsenophenylglycin and novoflavin, and the drug was injected into the jugular vein in a 1 per cent. solution in normal saline.

In the remaining cases salvarsan was injected at an early stage of infection, these results being free from any sources of error connected with treatment with other agents. The strain used had been recovered from a sheep dying after unsuccessful treatment with arsenophenylglycin after it had been passed through five untreated sheep. The drug was given in a 1 in 500 neutral solution.

By experimentation with ten sheep the dose was fixed at 0.04 g. per kilogramme.

In no instance was there a complete disappearance of trypanosomes from the blood.

Combinations of trypanocidal agents.

1. Arsenophenylglycin and Novoflavin.

Of four equines subjected to this treatment, three received the novoflavin after an interval of 52 days, when a relapse following treatment with arsenophenylglycin had already been demonstrated. In the other case an interval of nine days only separated the administrations of the drugs. In no case were trypanosomes again seen in the blood and inoculations were negative. The animals which had been in poor condition at the commencement of the experiment were killed after two months on account of debility.

Fourteen sheep were treated. Two died within a day from intoxication and two succumbed to accidental complications. In two instances in which the injections were given on successive days, no relapses were observed and the animals died from accidental causes nine and fourteen months after the completion of the experiment. The remaining sheep showed relapses.

Five of these sheep were given a second dose of novoflavin. Two died shortly after, two had relapses, and one survived with no relapse.

The two drugs were administered simultaneously to a horse which had resisted treatment with arsenophenylglycin, and to two sheep which had not been treated. The horse died of debility after 52 days without a return of trypanosomes being recorded, and the sheep died in 4 and 12 days without relapse.

Three sheep were killed by the simultaneous administration of 0.07 g. of arsenophenylglycin, 0.025 g. trypanblue, and 0.001 g. sodium arsenite per kilogramme body weight.

The following treatment was administered to five sheep:

0.04 g. arsenophenylglycin, 0.01 g. trypanblue, and 0.008 g. tartar emetic. These were given intravenously with intervals of two days, and were followed at an interval of 11 days by 0.05 g. arsenophenylglycin and 0.008 g. tartar emetic, these being given half subcutaneously and half intravenously.

In two sheep which received both treatments a cure appears to have been effected, and in the other three the first course of treatment appears to have been sufficient to effect a cure, no relapses having been noted up to 130 days.

It was found that larger doses could not be administered with safety.

A single dose of arsenophenylglycin followed by successive doses of emetic proved fatal in four instances.

Relapses followed in every case when the treatment consisted of alternate injections of 0.05 g. arsenophenylglycin and 0.01 g. of tartar emetic.

The simultaneous injection of these drugs was unsuccessful in every case.

A combined treatment consisting of quinine and atoxyl was also unsuccessful.

- (504) LAMBALLE (F. W.). Trypanosomiasis and Surra. A preliminary Note upon the effect of Pancreatic Enzymes upon the Trypanosome of Surra. With an Explanatory Note by J. Beard, D.Sc.—pp. 4. 1913. June 9. Edinburgh: Otto Schulze & Co.

In this brief note the author states that he administered to three relapse cases of surra in mules 3,750 units of trypsin and 1,000 units of amylopsin on two occasions at an interval of one day. At the time of treatment trypanosomes were numerous in the blood. On the day after the second administration only degenerated forms could be found in the blood, and on the following day no trypanosomes were to be found. The records are not carried any further.

- (505) OFFERMANN. Zur Frage der Immunität bei Trypanosomenkrankheiten. [Immunity against Trypanosomiasis.]—*Zeitschr. f. Veterinarkunde*. 1913. July. Vol. 25. No. 7. pp. 299-301.

A mare which was inoculated on four occasions with mouse or rat blood containing the trypanosome of dourine failed to show any symptoms of infection save that periodic rises of temperature were recorded. That the animal was actually infected was proved by repeated inoculations into mice. It was found that the blood was infective during the periods when the temperature was elevated, the inoculations failing during the afebrile periods. Agglutination and complement fixation tests both gave positive results.

- (506) DARLING (S. T.). The Immunisation of Large Animals to a Pathogenic Trypanosome [*Trypanosoma hippicum* (Darling)] by means of an Avirulent Strain.—*Jl. Experimental Med.* 1913. May. Vol. 17. No. 5. pp. 582-586.

A strain of this trypanosome that had survived in a guinea-pig for the exceptionally long period of 336 days was found on subinoculation on the 279th and 336th days to be feebly pathogenic when compared with other strains and with the same strain at an earlier period of infection. A dog inoculated on the day of death (336th) became infected but recovered. The usual duration of the infection in the guinea-pig is from one to four months. This strain was found to be of decreased virulence when used for the inoculation of a dog, a guinea-pig, and two mules.

In the case of one mule the period of incubation was lengthened to 11 days and the animal eventually recovered. This and other animals inoculated with a virulent strain were treated with arsenic, but the mule inoculated with the avirulent strain was the only one which survived. The animal passed through a typical attack and then appeared to recover. Its blood, however, remained infective for months, and it was only after rather more than a year after infection that it failed to infect a rat and a guinea-pig by inoculation, the dose used for the inoculation of the rat being 10 cc.

The history of the second mule infected (by means of *Musca domestica*) with the avirulent strain was very similar, but this animal was twice inoculated with a virulent strain without any infection being caused.

(507) BONGER (C.). Ueber die Morphologie und das Verhalten der von P. Behn in deutschen Rindern nachgewiesenen Trypanosomen bei künstlicher Infektion. [The Morphology of the Trypanosomes described by Behn as occurring in German Cattle in Animals infected experimentally.]—*Zeitschr. f. Hygiene und Infektionskrankh.* 1913. July 17. Vol. 75. No. 1. pp. 101-117. With 1 coloured plate.

The author has carried this trypanosome through four generations in calves. The course of the infection in each of the animals is given.

Three forms of the parasite are described:

a. The large slender type.—These measure on an average 55 microns in length by 2.5 in width. Their cytoplasm stains deeply, and the nucleus which is placed posterior to the centre of the body is generally round and compact. The blepharoplast as a rule lies nearer the nucleus than the posterior end, and is frequently kidney shaped.

b. The large broad type.—These measure about the same as the preceding in length, but are twice the width. They stain more faintly and their cytoplasm has a vacuolated appearance. The greatest breadth lies between the blepharoplast and the nucleus. The protoplasm frequently contains numbers of granules which stain of a deep violet tint. The nucleus is often placed transversely to the length of the parasite. The blepharoplast is about midway between the nucleus and the posterior end.

c. The small slender type.—These measure on an average 39 microns in length by 2 in width. The nucleus lies in the long axis of the body and the blepharoplast lies nearer to the posterior end of the body than to the nucleus. The blepharoplast is large and its shape often suggests that a piece has been cut out of it. The undulating membrane is rather poorly developed and the flagellum is fairly long.

The largest number of parasites were found in the liver, and the long slender forms outnumbered the other forms by two to one. As many as 50 parasites could be found in a single smear (whether "thick" or "thin" is not stated) made from the liver.

A few parasites were found which were rounded at the posterior end and there were a few round parasites. These measured on an average 9 microns in diameter.

The author failed to infect any animal other than the ox. The period of incubation was about 7 days. In one instance an inoculation proved successful when parasites had not been seen in the blood for 6 weeks.

Apart from a rise of temperature to about 40° C. which occurred in some cases before trypanosomes appeared in the blood there was no disturbance of health.

Parasites were discoverable in the blood for about 10 days.

- (508) PONSSELLE (A.). *Technique pour le Coloration de Trypanosomes et Trypanoplasmes de Culture.*—*Compt. Rend. Soc. Biol.* 1913. May 23. Vol. 74. No. 18. pp. 1072-1073.

1. The dried smears should be covered with the following mixture:—

Absolute alcohol 50 cc.

Tincture of Iodine (French Pharmacopoeia) 10 drops.

Allow this to act for 5 minutes, wash with absolute alcohol and allow the specimens to dry.

2. Cover the specimen with serum. Horse serum heated to 56° C. does very well. Leave for 5 minutes. Wash in distilled water.

3. Stain with Giemsa made up in the usual way for 15 to 30 minutes. Wash and dry.

The method is of especial value for the staining of cultures of trypanosomes and trypanoplasms, but it does not give better results than those obtained in the usual manner with blood containing parasites.

- (509) KERANDEL (J.). *Trypanosomes et Leucocytozoon observés chez des Oiseaux du Congo.* [Trypanosomes and Leucytozoa found in Birds in the Congo.] — *Ann. Inst. Pasteur.* 1913. June. Vol. 27. No. 6. pp. 421-439. With 2 coloured plates.

The present article appears to be an elaboration of one which appeared in the *Bulletin de la Société de Pathologie Exotique* in 1909.

The descriptions given are based upon the examination of preparations made as soon after the death of the hosts as possible, fixed in absolute alcohol and stained with Giemsa. In most cases opportunity did not offer for the examination of moist preparations.

Trypanosome of the Guinea-fowl (Numida meleagris).

This parasite has a long slender fusiform body which is drawn out at both ends, particularly at the posterior extremity. The cytoplasm stains of a deep bluish-violet colour at the centre of the body, but more faintly at the extremities. It is granular and shows some ill-defined granules.

The nucleus is rounded, centrally placed, and somewhat ill-defined.

The centrosome is oval and large, and is placed at some distance from the posterior extremity. The undulating membrane is well-developed and is thrown into voluminous folds. There is no free flagellum. The parasite measures about 70 microns in length and 4 microns at its greatest breadth.

Trypanosome of the Francolin (Francolinus bicalcaratus).

The author believes this to be a new species and suggests the name *T. francolini*. The special characters of the parasite are its great length as compared with its slenderness, and the great distance between the nucleus and the centrosome (nearly 20 microns).

The organism is drawn out at both ends. The cytoplasm stains deeply and is granular, and contains a number of vacuoles, one of which is elongated and starts at the centrosome and extends forwards. The ectoplasm shows discrete longitudinal markings. The nucleus is rounded and placed about the middle of the body. The centrosome is large and stains deeply. The undulating membrane is well-developed, but there is no free flagellum. The parasite measures from 55 to 65 microns in length and from 3 to 3.6 microns in width.

Trypanosome of Pycnonotus tricolor.

In the fresh state this parasite does not execute very rapid movements. The centrosome is very refractile and can be seen at that point in the body where it begins to taper. The nucleus is less clearly seen. The undulating membrane is very mobile and is thrown into a number of folds. Sometimes the movement involves translation but sometimes there is none. During movement the posterior portion of the body appears to be passive. When there is no movement of translation the parasite assumes a curved outline, and while revolving upon itself describes an almost perfect circle. In stained preparations the organism is observed to taper markedly towards its extremities, especially in the anterior direction.

The nucleus is rounded and placed towards the middle point of the body, but is sometimes a little posterior to that.

The centrosome is large and rounded, and lies in a large oval vacuole.

The parasite measures about 50 microns in length by 5.5 in width.

The trypanosome is said to differ from that described by MATHEIS and LÄGER in a bird belonging to the same family, in that the latter showed a rod-shaped centrosome, the membrane was thrown into a few folds, and the posterior extremity tapered to a less degree.

Trypanosome of the Vidua (Vidua serena).

This parasite somewhat resembles *T. bouffardi*, but the latter has a thicker body and a rod-like centrosome. The organism differs from *T. paddae* in that its centrosome is small and it shows no longitudinal markings.

Trypanosome of the Screech-owl (Strix flammea trimaculata).

Only one specimen of this parasite was found. It is possibly identical with *T. avium*. The length of the specimen found was 55 microns and the breadth 5 microns. There was a very slender free flagellum measuring 9 microns.

Trypanosome of the Roller (Eurystomus gularis).

This is a somewhat stout trypanosome and measures from 50 to 57 microns in length by 5 to 9 microns in width. The extremities do not taper much. The body stains deeply except posterior to the centrosome where it takes a very pale tint and it shows longitudinal striations. The centrosome is large. There appears to be no free flagellum in the majority of the organisms, although in three specimens a short one could be made out.

Trypanosome of the Fern-owl (Caprimulgus fovei).

Two types of trypanosome were found in this bird.

The larger trypanosome in the fresh state is very active and its movements always involve translation.

In stained preparations it is seen that the posterior extremity of body of the parasite terminates somewhat bluntly and suggests the head of a porpoise in shape, while anteriorly the body tapers off gradually from the nucleus. The whole of the body takes a deep blue colour including the portion posterior to the centrosome. The nucleus is clearly outlined and is placed rather posterior to the centre of the body. The centrosome is rod-shaped. The parasite measures from 43 to 47 microns in length by 5 to 6 in width.

The smaller trypanosome is not so active, and translatory movements are not observed. In the fresh state it can be seen that the undulating membrane makes three turns round the body in a spiral manner.

In stained preparations the body is faintly coloured. The extremities are seen to be rather bluntly conical. The centrosome is large and rounded, and situated quite close to the posterior extremity. The undulating membrane itself is scarcely visible, but the flagellum enables its course to be followed. No free flagellum can be seen.

The parasite appears to be a little more than 20 microns in length by about 5 microns in width.

In the blood of a bird about the size of a lark, the species of which was not determined, a trypanosome closely resembling the small parasite found in the fern-owl was discovered.

Leucocytozoa.

In the majority of cases these haematozoa were included in cells which in some cases were rounded and in others fusiform. The author agrees with MATHIS and LÉGER that the same parasite may be found in these two types of cell. He considers that the fusiform cells represent erythroblasts (cells related to erythrocytes but devoid of haemoglobin) and the rounded cells mononuclear leucocytes. The author's classification is based upon the view that each species of parasite infects only one family or allied genera.

Leucocytozoon of the Guinea-fowl (Numida meleagris).

This organism was found in the blood of the bird that was the host of the trypanosome already described. The parasite appeared to be practically identical with that described as occurring in the blood of a closely allied species in the Soudan, and it closely resembled that observed by MATHIS and LÉGER in domestic poultry in Tonkin.

In the fresh state the body of the parasite appears granular and the nucleus is not visible. In stained preparations the sexual forms are easily distinguished.

Macrogametes measure from 14 to 18 microns in length by 5 to 8 in width, and possess granular cytoplasm which stains of an intense blue. The nucleus is rounded, oval, or crescentic, and composed of compact chromatin granules.

The microgametes are slightly more slender than the macrogametes. The cytoplasm stains of a faint pink tint. The nucleus is large and ill-defined.

The host cell in both instances becomes drawn out at the poles, and in most cases contains numbers of red granules.

Leucocytozoon of the Francolin (*Francolinus bicalcaratus*).

The parasite found in this species was of the same type as that found in the Egyptian guinea-fowl, but on account of certain morphological characters the author believes it to be a distinct species.

The cytoplasm of the macrogametes is granular, vacuolated, and stains blue. The nucleus is small, oval, and contains only a small amount of chromatin.

The cytoplasm of the microgametes is more finely granular and stains pink or pale blue. The nucleus is large and oval, and appears as a pink vacuole placed transversely across the body. The microgametes are as a rule a little smaller than the macrogametes.

The host cells are sometimes leucocytes and sometimes erythroblasts.

Leucocytozoon of the Screech-owl (*Strix flammea trimaculata*).

The leucocytozoon found in this species closely resembles that found in the little owl and possibly the two are identical.

Leucocytozoon of the Fern-owl.

In this species parasites were found both in erythroblasts and in leucocytes. The parasites contained in the former were more elongated than those present in the latter.

The cytoplasm of the macrogamete is granular, vacuolated, and stains of an intense blue colour. The nucleus is large, poor in chromatin and stains faintly.

The microgametes possess only a small amount of cytoplasm which stains of a greyish-blue colour. The nucleus is very large and owing to the presence of small granules of chromatin stains of a pink colour.

The parasite differs from that described by DANILEWSKY in that it is larger, the nucleus stains more faintly, and there is more marked flattening of the nucleus of the host cell.

Leucocytozoon of the Roller (*Eurystomus gularis*).

The organisms were very scanty in the blood of this bird, a dozen only being found in two preparations which were thoroughly examined.

Like the preceding, this parasite is encountered in two types of host-cell.

The macrogametes have a granular protoplasm in which are scattered irregular vacuoles. The nucleus is oval and contains a large amount of chromatin.

The microgametes are smaller and stain of a faint pink colour. The nucleus is represented by a diffuse mass of finely granular chromatin. In the young parasites it forms the bulk of the organism.

SPIROCHAETOSIS.

- (510) MARCHOUX (E.) & COUVY (L.). Argas et Spirochètes. Premier Mémoire. Les Granules de Leishman.—*Ann. Inst. Pasteur*. 1913. June 25. Vol. 27. No. 6. pp. 450-480. With 15 text-figures.

The granules may be found in the tick in all stages from the egg to the adult. In the egg they are found scattered through the whole of the mass, but in the larvae they are localised in the Malpighian tubes, and in the adult in the ovules and genital passages also. The granules are always intracellular in position, and are collected together into little masses. In sections it can be seen that the entire protoplasm of the affected cells is packed with masses of small spherical bodies.

In the epithelial cells of the genital passages the granules are as a rule less numerous. In young ovules, before the formation of any vitelline, the granules can be seen arranged in more or less regular rows in the protoplasm. In this situation distinctly rod-like granules can be found. A similar disposition of the granules is somewhat rarely seen in the Malpighian canals.

It is stated that English authors have been mistaken in supposing that all spirochaetes disappear from ticks that are kept at a temperature of 15° C. to 28° C. for several weeks after an infective feed, this view being based upon the results of most careful microscopic examinations. The authors claim to have been able to find spirochaetes in the coelomic liquid of ticks that have been kept at 28° C. for 45 days. They claim that gentian violet is far more reliable for the demonstration of the organisms than Giemsa.

Starvation does not cause the disappearance of the spirochaetes. They have been able to find spirochaetes in the coelomic liquid of ten ticks which were starved and kept at a temperature of 15° C. for 11 months.

In a second series of experiments it was found that alternation of temperature between that of an ice chest and that of the laboratory failed to sterilise the coelomic liquid, and that spirochaetes were actually more numerous after five months than they were before.

Brief details are given of one of a series of experiments by which it was shown that the ingestion of antibody from a hyper-immunised fowl failed to kill the spirochaetes in ticks, organisms being found nine months later, although the ticks had had no infective feed during the interval. The serum of the fowl used was very active *in vitro*. One of the ticks used in this experiment laid a batch of eggs, and spirochaetes were found in the larvae when they hatched two months later.

The authors state that they have failed to sterilise ticks by the direct injection of active immune serum. The injection caused a temporary disappearance of the parasites, but if the ticks were given a feed of normal fowl blood parasites reappeared in a few days in 60 per cent. of instances.

In starved ticks the spirochaetes are smaller than normal and so slender that they may easily escape observation, but within

twenty minutes of a starved tick engorging normal blood the parasites can be seen to have increased in size up to the normal. If ticks are deprived of their organs of sense in the manner devised by HINDLE and are then allowed to engorge themselves with salt solution through a membrane, no enlargement of the spirochaetes is observed.

Experiment has shown that these slender spirochaetes are virulent by inoculation.

The following is a brief account of the authors' views regarding the fate of spirochaetes ingested by ticks from an infected fowl.

Some of the ingested parasites pass through the walls of the caeca and appear in the coelomic cavity about twenty minutes after the infective feed, but the great majority of the organisms remain in the caeca.

In the preparation of specimens of material from the interior of the intestine it was found to be easy to obtain intestinal contents without coelomic fluid by puncturing the dorsal carapace with a needle, the distended caecum becoming herniated through the opening, and thus allowing some of its contents to be withdrawn with a fine pipette. By practising this method the following facts were ascertained:—

Eighteen hours after engorgement the spirochaetes are still motile and present the same appearance as in the blood, but in a minority of them the chromatin is broken up into from four to eight rod-like fragments.

After twenty-four hours there is marked agglutination of the parasites, and with the ultra-microscope the agglutinated organisms appear as a series of refractile points. During this period of immotility it can be seen that the parasites act as centres of attraction for a number of exceedingly small refractile bodies which are present in the liquid. These granules collect along the organisms and give them a swollen appearance. A current in the liquid is sufficient to detach some of these granules, and unless the whole process has been observed the impression is created that the granules are being projected from the body of the spirochaete. When motility is completely lost the parasite becomes ill-defined and breaks up. This occurs in about a couple of hours.

After forty-eight hours the degeneration of the parasites is obvious. In many of them the endoplasm has become herniated through the ectoplasm at one or more places, either at the sides or ends of the organism.

After three days there may still be a few motile parasites, but these execute only a slow undulating movement. Many have by this time penetrated red corpuscles where they move around the nucleus. Numerous degenerated parasites and organisms with fragmented chromatin can be seen, and the whole preparation swarms with fine debris resulting from the degeneration of red corpuscles and spirochaetes.

After four days the haemolysis of the ingested blood is nearly complete. Spirochaetes that are abnormally slender and others that are abnormally short and stout can be observed in a state of active motility, while the organisms of normal dimensions are absolutely motionless.

By the following day there are but few intact corpuscles left, but abnormal spirochaetes are numerous and very motile.

The spirochaetes gradually disappear and they are rarely observed on the tenth day.

The authors disagree with those who see in the fragmented spirochaetes the stage preceding granule formation, and they consider that the fragmentation of the chromatin indicates the first stage in the degeneration of the parasite. Such forms are never seen in the circulating blood save during the period immediately preceding the crisis, when a few may be seen in the midst of agglutinated masses of the organisms. In the tick they are never seen external to the caeca and their presence there is limited to the period of digestion.

On the other hand this degeneration is observed whenever the spirochaetes are placed under adverse conditions. It can be seen in organisms contained in blood that is taken aseptically and kept in tubes, within 24 hours. The process is hastened if the tubes are kept at 37° C., but the degeneration is delayed for some days in the ice chest. When spirochaetes are placed in contact with antibody the degeneration is greatly accelerated. Exactly similar changes are said to occur in other blood-sucking parasites when these are engorged with blood containing spirochaetes.

That the short and stout spirochaetes result from degeneration or rather digestion is shown by the discovery of similar organisms in bugs and leeches engorged with blood containing spirochaetes, the authors having shown by experiment that spirochaetes present in the coelomic cavity do not pass into the caeca. Similar changes are observed in spirochaetes in tubes containing media which will not allow of multiplication of the organisms.

The nature of Leishman's granules.

It is said that the changes in shape observed under different conditions of temperature are also to be observed in ticks kept at the temperature of the laboratory.

The authors have never seen any transformation of the granules into spirochaetes, and they have not been able to induce such transformation *in vitro*. Further, if the granules develop into spirochaetes which subsequently again become changed into granules these latter should show the same susceptibility to specific antibodies as the spirochaetes themselves. This was not found to be the case.

In the majority of cases failure has followed attempts to produce infection by inoculation with Malpighian tubes containing the granules, nor does immunity always follow inoculation with such material.

If the granules are in reality a stage of the spirochaete, inoculation with enormous quantities should produce infection. This however does not occur in the majority of instances, and it has been found that the salivary glands which are devoid of granules are actually superior to the Malpighian tubes in their content of immunising material.

Attempts were made to produce granules in bugs and leeches by feeding them on blood containing spirochaetes, but in no case could any be found. In view of the fact that the bug and the leech are

species widely different from the tick the authors examined other acarine parasites and found that these also contained granules of similar shapes, sizes, and staining reactions, and in the same situations. The question as to what is the real nature of the granules of Leishman remains unanswered.

- (511) LAUNOY (L.) & LÉVY-BRUHL (M.). **Les Variations Numériques des Globules Blancs chez les Poules infectées de *Spirochaeta gallinarum*.**—*Compt. Rend. Soc. Biol.* 1913. April 18. Vol. 74. No. 13. pp. 754-756.

The numerical variations in the number of leucocytes were always very small, and much smaller than those observed in certain types of leucaemia in the fowl, and in fowls infected with tuberculosis.

There was an increase of polynuclears during infection, and an increase in the mononuclears after the crisis. The increase in the polynuclears occurred in two phases. In the first phase there was an increase in the polynuclear cells containing the rod-like granules, and in the second phase the polynuclear cells containing the rounded granules [eosinophiles?] were observed. The morphological variations although constant are not specific since they occur in other diseases.

LEISHMANIASIS.

- (512) GRAY (A. C. H.). **Leishmaniose naturelle du Chien à Tunis.**—*Arch. Inst. Pasteur Tunis.* 1913. No. 1-2. pp. 102-106.

From the figures published by different authors it appears that the number of infected dogs in the countries bordering on the Mediterranean is very variable. The highest percentage of infected dogs is 81 at Bordanaro (Messina) and the lowest 1.1 per cent. at Catano.

The author undertook the examination of the dogs in Tunis. This had already been done by other investigators and the figures arrived at have been in close correspondence, namely 1.6 to 1.8 per cent.

In view of the fact that the brothers SERGENT during July, August and September found 7.2 per cent. of dogs infected in Algiers, the possibility suggested itself that the percentage might be higher in Tunis during the later months of the year, the previous examinations having been carried out in the early part of the year. The author's examination was made during October and November, and out of 127 dogs only two were found to be infected, that is 1.6 per cent.—the figure arrived at by YAKIMOFF in 1911.

The materials used for examination are not stated.

- (513) PATTON (W. S.). **Is Kala Azar in Madras of Animal Origin? Preliminary Report.**—*Indian Jl. Med. Research.* 1913. July. Vol. 1. No. 1. pp. 185-195.

This paper contains the results of a number of animal inoculations with material taken from cases of the disease in the human subject and from experimentally infected animals, and the

results of the examination of a large number of bazaar dogs for the presence of leishmania.

In the first series of animal inoculations three monkeys, four dogs, two jackals, two guinea-pigs, two rabbits, two cats, four white rats, a goat, a calf, and a young pig were used. A fortnight later these animals were again inoculated with splenic emulsion from a case of the disease in the human subject. The material used was very rich in parasites.

In all the monkeys parasites were found in the organs and bone marrow. The animals had shewn wasting of the muscles with great weakness and there was marked diarrhoea.

Leishmania was found in all the dogs at the time of death and in three of them the infection was complicated with piroplasmosis. In those dogs in which piroplasms were found in the blood leishmania was also found, in many cases in leucocytes. The dogs lived several months after inoculation.

One of the jackals died with symptoms suggestive of kala azar, but a careful search failed to reveal the parasite, and a control jackal which had not been inoculated died showing the same symptoms. The other jackal that was inoculated not only contracted kala azar but also canine piroplasmosis. This is the first case that has come under the author's notice of a jackal which is ordinarily immune to canine piroplasmosis becoming infected.

Of the four rats, one became infected and died, and the other showed no parasites in its organs at the time of death.

The calf, pig and goat maintained perfect health.

In a further series of experiments five monkeys, ten white rats, and seven dogs were inoculated some with material from human cases and some with material from animals in the previous series.

One monkey which was inoculated subcutaneously with material from a monkey in the previous series became infected and died, but there was no evidence of a local lesion at the seat of inoculation. Parasites were plentiful in smears made from the organs. Another monkey died with symptoms suggestive of kala azar, but no parasites could be discovered at the post-mortem. The other three monkeys appear to have escaped infection.

Mention is made of five rats under these "additional experiments" and no evidence of infection was found in any of them, but it may be pointed out that according to their numbers two of those mentioned belong to the first series of experiments.

Two of the dogs inoculated respectively with fluid obtained by splenic puncture from a case of the disease in the human subject, and with bone marrow from the infected jackal, died but neither shewed leishmania on examination.

The two cats which were inoculated were killed six months later and both were found to be healthy.

Examination of spleen and bone marrow smears from bazaar dogs.

During the period January to November 1912, the author examined smears from the spleens of 1438 dogs destroyed in the lethal chambers and in no instance detected kala azar parasites.

It having been suggested that the parasites might be overlooked if the bone marrow were not examined, the author examined the bone marrow as well as the spleen pulp of 1,321 dogs, but failed to find a single parasite.

During the past seven years the author has examined about 300 cats, but has never seen leishmania in them.

The author feels justified in concluding "that the dog, and probably the cat, play no part in the transmission of kala azar in Madras."

(514) DONOVAN (C.). *Kala Azar: Its Distribution and the probable Modes of Infection.*—*Indian Jl. Med. Research.* 1913. July. Vol. 1. No. 1. pp. 177-184.

Geographical Distribution.—Outside India the presence of the disease has been recorded in China, the Yangtse Valley, and in the Soudan. "The disease as found along the littoral of the Mediterranean we may at present exclude and relegate to another form of Leishmaniasis, as this affects mainly, if not exclusively, children, and is apparently closely connected with the canine form of the disease."

In India the disease is found endemic in damp low-lying districts near the deltas of the Ganges and Brahmaputra. The disease has been said to occur in Assam, but the author suspects that the outbreaks were in reality of malarial origin, for the reason that the disease is said to have occurred as an epidemic and not endemically. The disease is endemic in the Madras Presidency.

Modes of infection.—There are two probable modes of infection:—(1) By means of insects, and (2) by ingestion, with infection though the mucous membrane of the intestinal tract.

1. By insects:—(a) Bed bugs.—Strong evidence of this has been adduced by PATTON, but several points are wanting to enable this view to be accepted in its entirety.

(b) *Conorrhinus*—*Conorrhinus rubrofasciatus* has been suspected by the author. He had not been able to get any multiplication of the organism in this species, but he thinks that it should be suspect for the reasons that it is known to suck human blood and that another species of the same genus has been found to transmit a human trypanosome in Brazil.

(c) Mosquitoes—Very little experimentation has been done with these diptera, and it is pointed out that they harbour flagellates which may be readily confused by an inexperienced observer with leishmania.

(d) House fly—*Musca nebulo* has been singled out as a transmitter, not by biting, but by transferring infective material to foodstuffs or to abrasions.

(e) Lice and ticks—These have been tested by PATTON with negative results.

(f) Dog-fleas—In Europe a fairly strong presumptive case has been made out as to the part the dog plays as an intermediary host, the dog flea being the actual transmitter.

"But the evidence adduced so far is not in all respects convincing. The occurrence of a natural flagellate of the flea has evidently not been taken into sufficient account."

The liver and spleen of a large number of dogs have been examined in India but no evidence of infection with leishmania has been obtained.

The author has been able to detect the parasite in the bone marrow of a dog inoculated into the liver with 3 cc. of blood taken from the spleen of a human being. In this instance examination of the liver and spleen failed to demonstrate the parasite.

2. By ingestion:—In a fairly large proportion of cases the disease in the early stages resembles typhoid, but the rise of temperature after a month or more of apyrexia identifies the nature of the infection. In all cases there are periods of diarrhoea and dysentery. In fatal cases evidence of ulceration of the large intestine is marked, and *Leishmania* is found in scrapings from these ulcers. This lends a certain amount of probability to the view that the parasite gains access to the system by way of the intestinal mucosa. It is true that *Leishmania* has not been found in the faeces of patients. Ankylostomes and trichomonads are frequent in such evacuations and the possibility is suggested that these might harbour the parasite. CHRISTOPHERS has examined ankylostomes but has not been able to find a trace of *Leishmania* in them.

(515) WENYON (C. M.). **A further Note on a Case of Dermal Leishmaniasis from South America, with the Results of Inoculation Experiments.**—*Jl. London School Trop. Med.* 1913. April. Vol. 2. No. 2. pp. 117-119.

In the first portion of this paper is given an account of the successful treatment of the disease with an ointment composed of equal parts of methylene blue, lanoline, and vaseline as recommended by CARDAMATIS and MELISSIDIS.

The second portion briefly summarises the results obtained by inoculating animals with cultures of the parasite. The cultures of the organism were much more luxuriant than those obtained from *L. tropica*. On two occasions intravenous inoculations were made into rabbits, and on one occasion a mouse was inoculated intraperitoneally, but in each case the result was negative. A human being inoculated by intracutaneous injection and by scarification failed to become infected.

A dog originally inoculated with material derived directly from the patient became infected and from it material was obtained for the successful inoculation of a second dog. From the second dog were inoculated a puppy—intravenously, two mice—intraperitoneally, and a cat on each ear. The puppy and the mice failed to become infected, but the cat developed two small pinhead nodules at the seats of inoculation after two months. *Leishmania* were discovered in the lesions both by culture and by microscopic examination.

- (516) KOHL-YAKIMOFF (Nina), YAKIMOFF (W. L.), & SCHOKHOR (N. J.). Leishmaniose Canine à Tashkent.—*Bull. Soc. Path. Exot.* 1913. June. Vol. 6. No. 6. pp. 432-433.

The authors have examined 76 dogs at Tashkent (Turkistan) and have found 22 affected with leishmaniasis.

TOXOPLASMOSIS.

- (517) LAVERAN (A.) & MARULLAZ (M.). Infections du Lapin par le *Toxoplasma gondii*. [Infection of the Rabbit with *Toxoplasma gondii*.]—*Bull. Soc. Path. Exot.* 1913. April. Vol. 6. No. 4. pp. 249-254.

After pointing out that NICOLLE and CONOR failed to infect rabbits with this organism, the authors pass on to describe their experiments in this connection.

The material used for the inoculations was the peritoneal exudate of a mouse infected with the parasite.

Eight rabbits were used. Of three inoculated into the peritoneum two showed no infection, but the third died on the eighth day of a generalised infection. Parasites were very scanty, especially in the peritoneal exudate.

Five rabbits were inoculated intravenously and all became infected, death occurring in from 4 to 7 days.

The authors state that the failure of NICOLLE and CONOR to infect rabbits was due to the fact that they used intraperitoneal inoculations only. Young rabbits are more susceptible than adults.

In young rabbits white nodules appear on the surface and in the substance of the liver, giving the organ a characteristic appearance.

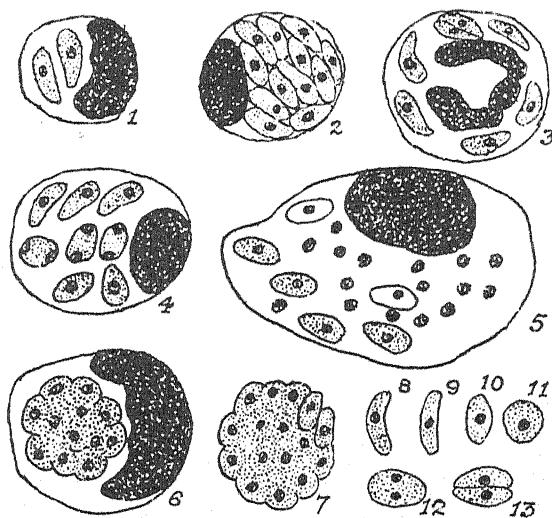
In rabbits inoculated intravenously the parasite may be present in fairly large numbers in the liver, and very frequently is found in the hepatic cells. Histologically the lesions are identical with those produced by *T. cuniculi*, and are due in all probability to the production by the parasite of a toxin which causes necrosis, and which is also, no doubt, the cause of death.

There is always enlargement of the spleen and the parasite may be present in large numbers. Microscopic examination reveals the presence of small centres of necrosis.

In the majority of rabbits dying of the infection there is congestion of the lungs, and parasites may be present in considerable numbers.

The organism is to be found in the bone marrow and in the brain, but it has been found in the blood of one rabbit only.

In one instance a rabbit inoculated intraperitoneally failed to show any signs of infection, and when inoculated intravenously it again resisted infection. Possibly in this instance the first inoculation caused a latent infection which produced immunity.



Toxoplasma gondii, endocellular or free in the peritoneal exudate of infected mice. (From the paper by LAVERAN & MARULLAZ in the *Compt. Rend. Acad. Sci.* 1913. Vol. 156. No. 17. pp. 1298-1302.)

1 and 2. Mononuclears containing toxoplasms. 3. Polynuclear containing toxoplasms. 4. Endothelial cell containing toxoplasms, three of which are dividing. 5. Endothelial cell containing toxoplasms, some with clear outline and others of which the nuclei only are seen. 6. Endothelial cell with agglomerated toxoplasms. 7. Agglomeration of toxoplasms; the contours of two are seen. 8-11. Free toxoplasms. 12-13. Division stages. (Magnification about 1,600 diameters.)

(518) LAVERAN (A.). *Présentation d'un Chien infecté de Toxoplasmosse.*—*Bull. Soc. Path. Exot.* 1913. May. Vol. 6. No. 5. p. 294.

At the meeting of the Society held on May 14th, 1913, Laveran shewed a dog that had been inoculated into the saphena vein with dilute peritoneal exudate obtained from a mouse infected with *Toxoplasma gondii*. After inoculation the dog rapidly lost flesh, and opacity of the cornea made its appearance in about three weeks. A fortnight after inoculation the parasite was found in the blood and there was marked increase in the number of polynuclear leucocytes. In one case a single leucocyte contained nine parasites.

The possibility is suggested that *T. cuniculi*, *T. gondii*, and *T. canis* are identical.

(519) SPLENDRE (A.). *Des Formes Flagellées et des Gamètes dans le Toxoplasma cuniculi.*—*Bull. Soc. Path. Exot.* 1913. May. Vol. 6. No. 5. pp. 318-323. With 1 plate.

In a previous communication the author suggested the possibility that there might be a flagellate stage in this parasite, basing the suggestion upon a few observations from which the possibility of error was not excluded. In the present paper a description is given of the flagellate form, the occurrence of which the author claims to have established.

The flagellate referred to was found in a preparation from the liver of a pigeon experimentally infected with *T. cuniculi*.

The body of the parasite measured 5.5 microns in length and 2.2 in width. It was slightly curved and had one extremity rounded and the other somewhat pointed. The protoplasm was vacuolated to some extent and stained pale blue. The nucleus was rounded or slightly elongated in the transverse diameter of the body, and situated at a point one third of the length of the body from the more pointed end. It appeared to be composed of a chromatic zone without obvious structure. There was a large caryosome from which started a filament. This passed along the convex border of the parasite and gaining the pointed extremity was continued for a distance of 14 microns as a free flagellum. This flagellum appeared to be composed of a number of rods of chromatin enclosed in a thin protoplasmic envelope. The terminal portion of the flagellum was a little swollen. The exact significance of the flagellated form is not quite clear, as in the author's opinion he has observed the true gametes of the parasite. These bodies he has observed in smears made from the pectoral muscles of experimentally infected pigeons. On the surface of these muscles are to be observed little greyish streaks suggestive of sarcosporidia. Microscopic examination of these shewed that they were collections of toxoplasma. The dividing forms which have already been recognised could be found, and in addition to these there were other forms presenting some slight differences. The principal difference presented by these forms was the situation of the nucleus, which was placed close to one extremity of the parasites. In stained preparations these forms with the displaced nucleus were sometimes as numerous or even more numerous than the dividing forms. The bodies were fusiform, and stained of a deep blue tint, and sometimes shewed blue granules. The nucleus was formed of a compact or granular rounded mass of chromatin.

Some of the preparations in which these bodies were found were made direct with the muscle juice, but in other cases the material had been first diluted with salt solution and the preparations were allowed to dry in the air. In the latter cases some of the bodies were found to have undergone changes of shape, the alteration being from the fusiform to the round, and also an increase in size. In these parasites the nucleus was generally close to one side of the body and at the opposite side of the organism there were one or two little rod-shaped pieces of chromatin, which are believed by the author to result from reduction of the nucleus.

In some of the parasites there were two or four nuclei resulting from division of the primary nucleus, and in some of these two of the nuclear fragments were disintegrating, while from the others were developed two chromatin filaments which in many cases passed right across the body of the parasite and projected free from the opposite side for a distance of three or four microns.

In preparations made direct from the tissues and fixed at once the fusiform parasites shewed no modifications whatever, and the

author therefore concludes that the production of the chromatin filaments must have taken place outside the body. The author suggests that the production of these forms in the superficial tissues indicates infection of an intermediate bearer, but states that no definite conclusion can be arrived at at the moment. It is suggested that *Stomoxys* is the intermediate host.

- (520) MARULLAZ (M.). Au Sujet d'un Toxoplasme des Oiseaux. [Toxoplasmosis of Birds.]—*Bull. Soc. Path. Exot.* 1913. May. Vol. 6. No. 5. pp. 323-326. With 9 text-figures.

After quoting at some length from a paper by Laveran which appeared in the *Comptes Rendus de la Société de Biologie* in 1900, the author gives a brief description of his examinations of a number of different species of birds.

Marullaz has found that the best results are obtained with preparations fixed in absolute alcohol for 4 to 6 hours and stained in Giemsa (1 drop to the cubic centimetre) for six to ten hours.

The majority of the investigations were carried out on *Padda oryzivora*.

The organisms, which often measure 5 or 6 microns in length by 2.5 to 3 in width, may be found in the liver, lung, and cardiac blood, but far more numerous in the spleen and bone marrow. In the blood the parasites are always within leucocytes. Multiplication takes place by longitudinal division.

The parasite has also been found in *Estrilda phoenicatis*, *Lagonosticta senegala*, *Quele aerythrope*, *Pyromelona franciscana*, and *Fringilla coelebs*.

Attempts to transmit the infection from padder to padder did not yield very satisfactory results, as it was impossible to determine whether a bird, which on post-mortem was found to be infected, had contracted the infection naturally or as a result of the inoculation, it being impracticable to examine the bone marrow before the experiment.

Attempts to transmit the infection to the pigeon by intravenous inoculation as a rule failed.

The author has been unable to cultivate the parasite on simplified Novy medium or in the peritoneal cavity of the white mouse.

The parasite appears to be distinct from *Toxoplasma cuniculi* and *Toxoplasma gondii* in that it is not inoculable either to the pigeon or to the mouse.

- (521) MESNIL (F.) & SARRAILHÉ (A.). Toxoplasmose Expérimentale de la Souris; Passage par les Muqueuses; Conservation du Virus dans le Cadavre.—*Compt. Rend. Soc. Biol.* 1913. June 27. Vol. 74. No. 23. pp. 1325-1327.

Having shown in a preliminary experiment with *T. gondii* that the deposition of a drop of peritoneal exudate from an infected mouse on the conjunctiva and in the vagina of other mice caused

infection, the authors carried out a number of experiments on the same lines with the following results:—

| Mucous membrane. | Results. | | Average duration of infection. |
|------------------------|-----------|-----------|--------------------------------|
| | Positive. | Negative. | |
| Vagina | 4 | 0 | 10 days. |
| Prepuce | 0 | 2 | — |
| Conjunctiva | 1 | 0 | 13½ days. |
| Vagina and conjunctiva | 2 | 0 | 10 days. |
| Mouth | 2 | 1 | 14½ days. |

Although positive results followed the placing of infective material on mucous membranes, it was found that infection would not take place through sound skin.

The duration of the disease was greater than that following intraperitoneal inoculation, and the dose administered did not appear to have much effect upon the result obtained. In every case the resulting infection was a generalised one. It is remarkable that parasites were numerous present in the thymus, and that the lymphatic glands were enlarged and contained parasites. The organisms were also present in the thyroid and parotid glands, in addition to the usual viscera.

It was found that the peritoneal fluid of a mouse which was kept in the laboratory remained virulent for 18 hours after death, and in one instance it remained infective for 30 hours.

(522) LAVERAN (A.) & MARULLAZ (M.). *Recherches Expérimentales sur le Toxoplasma gondii*.—*Bull. Soc. Path. Exot.* 1913. June. Vol. 6. No. 6. pp. 460-468.

Mice inoculated intraperitoneally die in about five days, and it is easy to keep a strain running by subinoculations with peritoneal exudate every three days. The parasites are more numerous when the exudate is small in amount and viscous than when it is clear and serous.

There is enlargement of the spleen and congestion of the lungs. The parasite may be found in the liver and spleen, lungs, and, in some cases, in the bone marrow.

Grey mice and field mice can also be infected.

The authors failed to infect white rats either by intravenous or intraperitoneal inoculation, this result agreeing with that obtained by NICOLLE and CONOR. A dormouse also failed to become infected as a result of intraperitoneal inoculation.

Rabbits can be infected either by subcutaneous, intravenous, or intraperitoneal inoculation. Three out of six rabbits inoculated into the peritoneum failed to become infected, and they resisted subsequent inoculations by the subcutaneous, intraperitoneal, and intravenous paths.

In the rabbit there is always wasting, and there is as a rule an abundant peritoneal exudate which is poor in parasites. The naked-eye appearance of the liver and the histology of the alterations produced are identical with those produced by *T. cuniculi*.

Parasites are generally numerous in smears from the congested portions of the lungs, but rare in the bone marrow.

Three young guinea-pigs were successfully infected by intraperitoneal inoculation from a mouse, and died in about 4 days. In addition to these animals the authors have succeeded in infecting the mole, hedgehog, shrewmouse, dog, pigeon and *Padda oryzivora*. They failed to set up infection in the rat, dormouse, fowl, several exotic passerines, frogs, and lizards.

The results of the experiments detailed in this paper confirm the opinion previously expressed by the authors that *T. gondii* and *T. cuniculi* are identical. The two organisms possess the peculiar characteristic of being inoculable to both birds and mammals, and yet the rat is refractory while the mouse is susceptible.

MESNIL states that the rat is refractory to intravenous inoculation, but that he has succeeded in inoculating a young dog intravenously. The authors have also discovered that *Toxoplasma gondii* is capable of penetrating healthy mucous membranes, particularly that of the vagina. It is capable of setting up a very severe inflammation in the vaginal mucous membrane in the mouse, and causing the production of a cellular and viscous exudate which contains numbers of living virulent toxoplasms.

523) NICOLLE (C.) & CONOR (Marthe). *La Toxoplasma du Gondi. Memoire Complet.* — *Arch. Inst. Pasteur Tunis.* 1913. No. 1-2. pp. 106-115.

Natural infection in the gondi.—As a rule no symptoms are observed save during the few hours before death. The animal is generally fat. There is a clear exudate in both pleural cavities, associated with congestion or hepatisation of the lungs. The spleen may be greatly enlarged, and, less constantly, there is slight enlargement of the liver. In rare cases there are no visceral lesions.

The distribution of the parasite.—The spleen as a rule contains immense numbers of the organism. Next in order stand the liver and the lungs. Parasites are rarely encountered in the kidneys and seldom or never in the blood or bone marrow.

The parasite varies in shape, being curved, pear-shaped, oval or round. The measurements vary from 2.5 to 6 microns in length by 2 to 3 in width. There is a single nucleus which is usually centrally placed. If it be not central that extremity of the parasite towards which it is displaced appears swollen, the opposite end being attenuated.

The parasite may occur free or enclosed in cells. These are generally mononuclear leucocytes, but they may be found in polynuclears.

Multiplication takes place so far as is known by longitudinal division only, and any number of daughter organisms up to 20 or 30 may be formed. These generally cling together forming a rounded mass and suggesting encystment, but no true encystment has been observed.

Experimental infection.—The infection has been transmitted to healthy gondis by intraperitoneal inoculation with bone marrow

from a naturally infected gondi, the material used being very poor in parasites.

Next to the gondi, the mouse is the most susceptible animal. Death after intraperitoneal inoculation generally occurs in 5 or 6 days. As a rule there is no generalised infection, but a local multiplication of the organism with the passage of a few parasites into the liver and spleen. At the post-mortem there is found an abundant greyish mucoid exudate in the peritoneal cavity, and the viscera are covered with a delicate false membrane. The parasite is encountered in large numbers in this exudate, enclosed in cells, and it is also found in the endothelial cells of the peritoneal membrane. Parasites are very scanty in the spleen, exceptionally present in the liver, and absent from the blood and bone marrow. In a few cases the infection has been successfully transmitted by intramuscular inoculation.

The guinea-pig is less certainly infected than the mouse by intraperitoneal inoculation, but when infection occurs the course of the disease and the lesions produced closely resemble those found in the mouse.

The pigeon is susceptible, but not so certainly as the mouse. The course of the infection is slower in the pigeon than in the mouse, and there is a tendency to generalisation. The sole method of infection which succeeds is intraperitoneal inoculation. Death takes place in from 9 to 30 days, and the most marked symptom is excessive loss of weight.

At the post-mortem there may or may not be found false membranes. The liver is enlarged and very friable. There is no enlargement of the spleen, but the pulp is very soft. The liver is found to contain the largest number of parasites, and after that the spleen, lungs and bone marrow. Parasites may be fairly numerous in the blood and kidneys. The parasite is not found in the false membranes except in those cases in which death takes place early.

Five attempts to infect young rabbits all failed, and entirely negative results have followed the inoculation of white rats, a young dog, two macaques, two toads, and two frogs.

The details of the experiments upon which the conclusions are based are given.

HORSE SICKNESS.

- (524) KUHN (P.). Die Immunisierung von Pferden gegen Pferdes-
terbe mit Hilfe von erhitzten Virus. [The Immunisation of
Horses against Horse Sickness with Heated Virus.]—*Zeitschr.
f. Immunitätsforsch. u. exp. Therap.* 1 Teil. Orig. 1913.
July 26. Vol. 18. No. 6. pp. 591-615.

In this paper the author records in detail the results of 25 experiments in which attempts were made to immunise horses against horse sickness by means of infective blood heated to 60° C. for varying periods. In some instances the virus was subjected to heat once only, but in other cases the heating was repeated a number of times. Other variable factors in the

experiments were, the dose used and the manner in which the material was administered. The virus used was either pericardial fluid or oedematous liquid from the lung.

In the first experiment 10 cc. of oedematous fluid from the lung was heated for two hours on three occasions and injected subcutaneously. The period of incubation was nine days and death took place on the fourteenth day.

An animal which was injected with 10 cc. of virus which had been heated 5 times for 2 hours and 20 minutes each failed to shew any symptoms, but when the animal was inoculated on the 26th day with 10 cc. of virus which had been heated three times only symptoms appeared and death occurred on the ninth day.

In a large number of the experiments the virus was heated to 60° C. for 2 hours and 20 minutes on four occasions, and of this material doses varying from 5 to 50 cc. were given, the dose being repeated on from 2 to 7 consecutive days.

It would appear from the table given that in eight or nine cases the heated virus was responsible for the death of the animals from horse sickness. In three or four cases the animals did not react in any way to the heated virus, but their immunity does not appear to have been tested with unheated virus. In 5 instances animals which received two or three doses of heated virus—generally one dose of about 5 cc. and one of about 40 cc. at an interval of a couple of days—resisted inoculation with 0.1 to 0.3 cc. of unheated virus. In the great majority of these cases the inoculations were intravenous. The author's conclusions are as follows:—

1. Inoculation with the virus of horse sickness frequently sets up reactions the cause of which has not yet been explained.
2. When oedematous fluid from the lung taken from a horse dead of horse sickness is heated to 60° C. the duration of the heating influences its virulence as indicated by a prolongation of the period of incubation.
3. The period of incubation may be as long as 45 days. Consequently in experiments with horse sickness a period of 45 days must be allowed to elapse before an animal is used for a new experiment.
4. When the virus is heated to 60° C. on four consecutive days the inoculation of 5 to 10 cc. subcutaneously in some instances causes death from horse sickness, but in other cases produces no illness.
5. When intravenous inoculations of virus treated in this way are given repeatedly at intervals of a few days, and to the total quantity of about 30 to 40 cc., a marked degree of immunity is established, and this by the 14th day after the first inoculation. Further experiments are required to ascertain the practical value of the method.

HELMINTHS.

- (525) SEURAT (L. G.). *Sur deux Spiroptères de Chat Ganté (Felis ocreata Gmel.)*—*Compt. Rend. Soc. Biol.* 1913. April 11. Vol. 74. No. 12. pp. 676-679. With 7 figures.

Spiroptera subaequalis Molin. This worm inhabits tumours in the pyloric portion of the stomach. It is distinguishable from *Spirocera sanguinolenta* of the dog by its smaller size (20 mm.) the conformation of the teeth in the oral cavity, the ovijector, and the much smaller size of the tail in the female.

The mouth is limited by a rim which is slightly indented at six places and it opens into a large buccal cavity attached to the walls of which are six very prominent teeth.

The ovjector resembles that seen in the spiroptera of the dog. It shows a pyriform vestibule with a long neck in which the sphincter projects obliquely, but while in the parasite of the dog the vestibule is lined with a thin chitinous layer and is packed with eggs, that found in *Spiroptera subaequalis* is far more narrow and is lined by a very thick chitinous membrane. The eggs seldom number more than two or three.

The worm is red in colour. There are two lateral papillae situated just behind the nerve ring. The excretory pore is ventral and below the papillae. There are no alae.

The oesophagus measures one quarter of the total length in the male and one-third in the female.

The female is from 19 to 21 mm. in length. The tail is short and obtuse. The vulva is not prominent and is situated 5 mm. from the cephalic extremity.

The male is a little larger than the female. The lateral wings of the caudal pouch are supported by four pairs of pre-anal rays and two pairs of post-anal rays. There is a single ray in front of the cloaca, and a group of eight small papillae which are difficult to see at the caudal extremity. The spicules are very unequal in length, measuring 2.25 mm. and 470 microns. There is one accessory piece.

The second spiroptera belongs to the genus Habronema, is of a red colour, but is found free in the stomach.

There are two very large alae which arise just in front of the nerve ring and extend for two-thirds of the length of the body. There are two cervical papillae. That on the left is situated in front of corresponding ala, and that on the right behind the nerve ring on the ala. [From an examination of the diagram given it would appear that these terms have been transposed.]

The mouth is surrounded by two lips which are markedly indented on their free border. The lips each show three teeth, the middle one of which is the largest and extends the whole length of the buccal cavity. There are also a dorsal and a ventral tooth which extend the whole length of the cavity, and, when viewed from in front, the mouth appears to be surrounded by eight teeth.

The oesophagus is divisible into three parts: an anterior portion where the muscular tissue is very strongly developed, a middle portion which is surrounded by the nerve ring, and a posterior glandular portion.

The female measures 8.5 to 9 mm. The excretory pore is 210 microns from the anterior extremity. The tail is regularly attenuated. The vulva is slightly salient and is situated just posterior to the middle of the body. The eggs show a little projection at one pole. The parasite is ovo-viviparous.

The male measures from 3.5 to 5.5 mm. The cloaca which opens between two prominent lips is placed 180 microns from the caudal extremity. There are four pairs of pre-anal papillae which are grouped in pairs, and two pairs of post-anal papillae.

The right spicule measures 280 microns, and the left 480 microns. There is one accessory piece.

- (526) RAILLIET (A.) & HENRY (A.). *Un Haemostrongylus des Bronches du Léopard*.—*Bull. Soc. Path. Exot.* 1913. June. Vol. 6. No. 6. pp. 451-454. With 2 text-figures.

The parasite described in this paper was found in the bronchi of a leopard killed at Dongou in the Belgian Congo.

The body of the worm is of a greyish colour and tapers slightly at both ends. The alimentary tract is of a darker tint and is entwined with the genital organs. The cuticle is striated longitudinally, and in the female there are at places delicate transverse folds.

The mouth is surrounded by six small papillae. The oesophagus is only slightly dilated at the posterior part and measures from 290 to 335 microns in length. The nerve collar and excretory pore are situated a little in front of the middle point of the oesophagus.

The male is from 10.5 to 12.4 mm. in length, and from 260 to 290 microns in thickness. The posterior rays of the caudal pouch are very short and are inserted into a wide main trunk. The middle and anterior rays are cleft. There are two long slender spicules which are striated transversely. There is no accessory piece.

The female measures from 20 to 23 mm. in length and from 460 to 525 microns in width. The tail which is bluntly conical has, at a point 35 to 45 microns from its extremity, two small lateral papillae. The anus opens 190 to 250 microns from the posterior extremity, and the vulva a little posterior to the middle point of the body. The uterus contains eggs in all stages of development. Eggs containing embryos measure about 80 microns in length by 50 in breadth. The embryos have a small projecting ventral lip at the anterior extremity, and a flexuous appendage at the caudal extremity. They measure from 250 to 270 microns in length and 12 to 15 in thickness.

The parasite is viviparous.

The authors conclude that the parasite belongs to the family *Strongylidae* and to the sub-family *Metastrongylinae*. The worms differ in characters of secondary importance only from *Haemostrongylus vasorum*, the parasite of the right heart and pulmonary artery of the dog.

On this account, and in spite of the fact that the habitat of the organism is the bronchial tubes, the authors propose to place it in the genus *Haemostrongylus* and call it *H. subcrenatus* in reference to the transverse folds in the cuticle.

MISCELLANEOUS.

- (527) MOLDOVAN (J.). *Sur le Développement du Leucocytozoon ziemanni (Laveran)*. Note préliminaire.—*Bull. Soc. Path. Exot.* 1913. June. Vol. 6. No. 6. pp. 428-429.

In two owls which died natural deaths the author has discovered organisms belonging to the asexual cycle of this parasite. The birds were at the same time hosts of *Halteridium noctuae*.

The preparations were made from the lungs, heart, brain, liver, kidneys, and bone marrow. In addition to the typical gametes there were found organisms included within lymphocytes and erythroblasts, which from their characters must be considered as young schizonts of the leucocytozoon. In some of the preparations, particularly those from the lungs and the brain, all stages of schizogony were found from the young schizonts up to those in which there were thirty nuclei or more.

In a general way the young parasites resembled those described by KEYSSELITZ and MAYER in the leucocytozoon of *Guttera pucherani*, and by von PROWAZEK in Sumatra fowls.

In some of the young parasites there was enlargement without division of the nucleus, and these shewed differentiations of their nuclei and cytoplasm suggesting phases in the development of gametes. Some forms were observed suggesting the stages of the development of the typical fusiform gametes.

The author states that in the same smears there were trypanosomes which were well differentiated into the male and female forms corresponding to the motile stages in the evolution of the gametes as described by SCHAUDINN.

It is said that the objection that might be raised that the forms observed were in reality stages in the development of *H. noctuae* cannot be upheld when the form and structure of the invaded cells and the resemblance shewn to the forms described by KEYSSELITZ, MAYER, and von PROWAZEK are taken into consideration.

The schizogony is said not to correspond with that described by FANTHAM for *Leucocytozoon lovati*.

(528) LÉGER (A.). Parasite des Hématies, Genre *Grahamella* (Brumpt), de *Mus maurus* (Gray). [A Parasite of the Genus *Grahamella* (Brumpt) in the Blood Corpuscles of *Mus maurus* (Gray).]—*Bull. Soc. Path. Exot.* 1913. April. Vol. 6. No. 4. pp. 247-249.

The author has found in the blood of *Mus maurus* at Bamako the rod-like organisms previously described by a number of authors as occurring in the blood of different animals, including the mole, gerbil, dormouse, and field-mouse. The organisms were found in 28 out of 125 rats examined.

There is not any distortion of the invaded corpuscles, and the number of organisms found in a single cell may vary from 6 to 80, but as a rule there are from 30 upwards. Invaded corpuscles are generally scanty in the blood taken from the tail, but more numerous in the heart blood. The organisms measure 1 micron in length, by 0.25 micron in width.

Good results may be obtained by staining with a mixture of Giemsa and acetone in equal parts, Giemsa (1 to 10) for fifteen minutes with slight differentiation with absolute alcohol, and with thionine-picric acid method devised by Sabrazès.

Under a high magnification some of the organisms appear to be attenuated in the middle as if in a state of transverse division. Rats, guinea-pigs, and rabbits have been inoculated in various ways without success.

- (529) ARCHIBALD (R. G.). *Aspergilliosis in the Sudan Ostrich*.—*Jl. Comp. Path. & Therapeut.* 1913. June. Vol. 26. No. 2. pp. 171-173. With 3 text-figures.

The lesions described in this paper were encountered in an ostrich in western Kordofan which had died with signs of marked emaciation. The disease principally involved the bronchioles. The upper portion of the bronchus was lined with dark granular masses projecting into the lumen. These masses were somewhat friable, but closely adherent to the wall. On the surface of the bronchiole adjacent to the lung tissue there were several "plaques" varying in size from a millet seed to a pea. The majority of them were discrete, but a few had coalesced. Their surface was marked with ridges and depressions.

In sections of the dark granular masses stained with Gram numerous filaments and conidia were found. Some of the filaments measured 6 microns in width and possessed very thick walls. Their extremities were dilated into club-shaped bodies measuring 18 microns in diameter, and upon these were arranged in radial fashion numerous sterigmata cells. At the distal end of each sterigmata cell were found numerous conidia measuring 2 to 3 microns, and there were large numbers of free conidia. Some of these cells had clear contents, others presented a dark-stained area in their centre, while others were filled with a mass of substance which stained densely with iron haematoxylin. The branching of the filaments was irregular, and some shewed very definite segmentation.

The "plaques" consisted for the most part of dense fibrous tissue with granular amorphous areas devoid of cellular elements. Scattered through the "plaques" were filaments 4 microns in breadth, which offered considerable resistance to staining.

Cultivation experiments with the object of ascertaining the species of the parasite could not be undertaken, but morphologically it was not unlike *Aspergillus fumigatus*.

- (530) KNOWLES (R.) & ACTON (H. W.). *A Note on Kurloff Bodies*.—*Indian Jl. Med. Research.* 1913. July. Vol. 1. No. 1. pp. 206-211. With 2 plates.

The authors have made series of blood films from a single guinea-pig, and have fixed and stained them in various ways, comparing the pictures obtained by the different methods.

Films were fixed in the following manners:—

1. air dry and fixed with methyl alcohol;
2. fixed while wet with methyl alcohol;
3. fixed while wet with formol-alcohol (formalin 1: absolute alcohol 9);
4. fixed while wet with Schaudinn's granule fixative (saturated aqueous perchloride 60 cc., absolute alcohol 30 cc., glacial acetic acid 3 cc.);
5. fixed while wet by plunging immediately into Schaudinn's fixative, kept at 60° C. on a water bath (saturated solution of perchloride in saline 2 parts: absolute alcohol 1 part).

Some of each of these were stained with Leishman's stain, Giemsa's stain, the original Romanowsky, and Mallory's iron-haematoxylin with watery eosin as a counter stain.

They have also made examinations by Ross's jelly method, the preparations being examined at room temperature, after incubation at 37° C. for one hour, and on a warm stage at 40° C.

Preparations have been examined under dark ground illumination and with vital staining with dahlia.

Their conclusions are as follows:—

"1. No reliance can be placed upon the appearance of the Kurloff bodies as seen in fixed films, unless the action of the fixative fluids has been previously investigated in order to see what fixation images are obtained. Otherwise the different fixation images obtained may be readily mistaken for stages in a cycle of development.

"2. In Kurloff bodies stained by a dahlia solution and examined on a warm stage we have been unable to trace any cycle of development.

"3. It seems somewhat unlikely—though not impossible—that a structure seen in the blood of 71 per cent. of apparently perfectly healthy guinea-pigs is parasitic in origin.

"4. We adhere to the view that they are cell inclusions of a non-parasitic nature.

"5. We believe that the Kurloff body is a vesicular structure, and probably of cytoplasmic origin. The guinea-pig is an animal in which these cytoplasmic structures are particularly well developed, e.g., the large archoplasmic vesicles in the testicular cells.

"6. In the course of the repeated examinations of the blood of pigeons in connection with the study of the *Halteridium* parasite we have occasionally seen somewhat similar bodies in the large mononuclear leucocytes and we doubt if the Kurloff body is specific for the guinea-pig."

(531) BOUILLIEZ (M.). *Nouvelles Recherches Expérimentales sur un Plasmodium des Singes.*—*Compt. Rend. Soc. Biol.* 1913. May 23. Vol. 74. No. 18. pp. 1070-1072.

The author has succeeded in inoculating the following monkeys with *Plasmodium inui*:—*Macacus nemestrinus*, *Cercopithecus cephus*, and *Cercopithecus callitrichus*. In the case of the first two the infection took a chronic course, but in the case of the last death occurred with haemoglobinuria on the twelfth day after inoculation. A male and female chimpanzee failed to become infected.

In two instances splenectomy was performed after the second appearance of the parasites in the blood. In one case the animal died as a result of peritonitis and in the other case the effect of the operation appeared to be to lengthen the period for which the parasites were present in the blood, but the animal died.

The results obtained in two experiments indicated that quinine is valuable as a preventive, but experiments designed to ascertain the curative value of the drug did not allow any definite conclusions to be drawn.

Attempts to cultivate the parasite by Bass's method at 40° C. failed, but in tubes containing defibrinated blood and dextrose there appeared to be a multiplication of the parasites when the tubes were kept at about 22° C.

(532) GRAYBILL (H. W.). The Action of Arsenical Dips in protecting Cattle from Infestation with Ticks.—*U.S. Dept. Agriculture. Bureau of Animal Industry. Bulletin 167. 1913. April 15. pp. 27.*

This bulletin contains details of a number of experiments carried out with the object of ascertaining the effects produced by oily substances upon ticks as well as experiments connected with arsenical dips. The following is the summary :—

"In this bulletin the factors entering into the efficacy of dips used against ticks are discussed. Dips act both in a direct destructive way and in a protective manner preventing infestation. The protective action of a dip may be in the nature of a destructive or of a repellant action. The influence of dips on oviposition and the viability of the eggs is a factor in efficacy.

"The ingredients of home-made arsenical dips and the known or probable functions of each ingredient are discussed.

"It is suggested herein that any protective action that the usual arsenical dips might have would be expected to be due to a toxic rather than to a repellant action. Watkins-Pitchford has shown that cattle dipped in arsenic are poisonous to ticks.

"Ticks are destroyed by dips either by suffocation or by poisoning, or by both means. Tests were conducted showing that ticks are suffocated by the closing of their respiratory openings (spiracles). It was found that practically all engorged females that had their spiracles closed with Canada balsam died. In other tests of the same sort, in which oils were used, Beaumont oil proved much less effective than Canada balsam, and cottonseed oil was practically without effect.

"Smearing the scutum and mouth parts of engorged females with oils and viscous substances had no influence on the mortality or oviposition, or on the per cent. of eggs hatching.

"In tests in which engorged females were dipped in Beaumont oil and in cottonseed oil the former proved very much more effective than the latter, and this is due in all probability to a toxic action. Beaumont oil had a marked influence on oviposition, on the number of eggs deposited, and on the viability of the eggs, whereas cottonseed oil has no effect.

"The possible avenues for the entrance of arsenic into the bodies of ticks are enumerated and the porose areas are pointed out as possible vulnerable points in the armour of the tick.

"Three cattle-dipping experiments were conducted with an arsenical dip containing 8 pounds of arsenic trioxide to 500 gallons of water, in order to test its protective action against tick infestation. Seed ticks were placed on the cattle at varying periods after they were dipped. In the first experiment the ticks were placed on the cattle at periods ranging from a few hours to four weeks, in the second from a few hours to two days, and in the third at five days after dipping. It was found that the dip rendered no protection when the exposure to infestation was five days or longer after dipping. The limit of protection ascertained in the experiments was two days. No tests were made covering the intervening period between two and five days.

"Arsenical poisoning which occurred among the animals in one experiment was apparently caused by undissolved arsenic in the dip. It would therefore seem that undissolved arsenic in a dip is highly dangerous.

"It is shown conclusively that the protective action of arsenic is dependent on its toxic action and not on a repellant action.

"As a result of incidental observations made on engorged female ticks from animals suffering from Texas fever it was found that the mortality of such ticks may be very high, as much as 95 per cent. The cause of this is not known. It may be nutritional in character, due to the change or impoverished condition of the blood absorbed, or it may be due to the parasitism of *Piroplasma bigeminum*, the micro-organism which is the direct cause of Texas fever.

"Observations made for the purpose of determining whether there was any relationship between the degree of infestation and the time elapsing

between the last dipping and the infestation, and also on the mortality of engorged females from dipped animals infested subsequently to dipping as compared with that of ticks from undipped animals were inconclusive. Oviposition and viability of the eggs of these ticks appeared to be unaffected."

BOOK REVIEWS.

- (533) BESSON (A.). *Practical Bacteriology, Microbiology and Serum Therapy (Medical and Veterinary). A Text Book for Laboratory Use.* (Translation from the 5th French edition by H. J. HUTCHENS.)—xxx. + 892 pp. With 416 illustrations. 1913. London: Longmans, Green and Co. [36s. net.]

The principal value of this book appears to lie in that portion of it which is devoted to laboratory technique, at least as far as the veterinary surgeon or student is concerned. The descriptions given of the organisms causing disease in the lower animals and of the diseases themselves are in some cases somewhat incomplete, and in some instances not quite accurate. In the description of the methods of infection in black quarter the following statements, which appear to be difficult to reconcile, occur within a few lines of each other: "The same dose of virus which will kill an ox when inoculated into the cellular tissues of the body will set up merely a benign infection if inoculated into the connective tissue of the neck," and "The inoculation of a virulent virus into the veins of an ox merely leads to a temporary rise of temperature; but if at the time of inoculation a trace of virus gains access to the perivascular tissues a fatal septicaemia ensues." Apart from the contradiction contained in these sentences it may be said that it is exceptional for the bacillus to multiply in the blood stream to such an extent as to warrant the term septicaemia being applied to the condition produced.

In connection with anthrax it may be noted that the bacillus is said to be capable of sporulating in a buried carcase, a view which has much to be said against it.

The pathogenic protozoa of the lower animals are very briefly dealt with and in some instances very important information is not given at all. For example, no mention is made regarding "blue bodies" in East Coast Fever. The error regarding the development of the sporoblasts in *Coccidium cuniculi* is perpetuated in that it is stated that the contents of the oocyst first divide into two and then into four sporoblasts, whereas the division into the four sporoblasts takes place simultaneously.

No mention is made of such important diseases as Johne's disease and contagious abortion.

For the most part the book is fully and excellently illustrated and the work of the translator and the publisher has been well done.

The scarcity of books dealing with the subjects of this volume makes the book a valuable addition to the literature devoted to them and it will on this account be welcomed by teachers and students.

- (533a) SCOTT (W.). **Clinical Bacteriology and Vaccine Therapy for Veterinary Surgeons.** 1st Edition. xii. + 222 pp. With 12 plates and 37 text-figs.—1913. London: Baillière, Tindall & Cox. [7s. 6d. net.]

The numerous errors and mis-statements of fact contained in this volume indicate that the author's knowledge of his subject is very limited. The book also appears to have been hastily and carelessly put together, as in many instances sentences are so constructed as to convey a meaning quite different from what was obviously intended. For example, on page 140 the following sentence appears:—"An animal injected with an immune serum lasts from three to four weeks." Well known names such as SCLAVO and SOBERNHEIM are misspelled. An administration is said to be made "per oram." Such examples could be given in abundance.

The book cannot be recommended.

RECENT LITERATURE.

[Continued from *Bulletin* No. 4, pp. 256-258.]**Babesiasis (Piroplasmosis).**

- (534) ZIEMANN (H.). Kurze Bemerkung zu dem Aufsatz: Ueber die Vermehrung von *Piroplasma canis* in Vitro von Knuth und Richters. [A Brief Observation regarding Knuth and Richter's Article on the Multiplication of *Piroplasma canis* in vitro.]—*Berlin. Tierärztl. Wochenschr.*, 1913. April 17. Vol. 29. No. 16, pp. 290-291.

Foot-and-Mouth Disease.

- (535) BÖHM (J.). Zur Pathogenese der Maul- und Klauenseuche. [The Pathogenesis of Foot - and - Mouth Disease.]—*Deut. Tierärztl. Wochenschr.*, 1913. May 31. Vol. 21. No. 22, pp. 337-338.

Lamziekte.

- (536) BURTT-DAVY (J.). Botanical Investigations into Gal-lamziekte.—*Second Report of the Director of Veterinary Research. Union of South Africa.* Oct., 1912. pp. 181-221. With 33 plates. (1913. Cape Town: Cape Times, Ltd., Government Printers.)
- (537) MITCHELL (D. T.). Lamziekte. *Second Report of the Director of Veterinary Research. Union of South Africa.* Oct., 1912. pp. 161-180. (1913. Cape Town: Cape Times, Ltd., Government Printers.)
- (538) THEILER (A.). Facts and Theories about Stijfziekte and Lamziekte.—*Second Report of the Director of Veterinary Research. Union of South Africa.* Oct., 1912. pp. 7-78. With 11 plates. (1913. Cape Town: Cape Times, Ltd., Government Printers.)
- (539) WALKER (J.). Investigations into the Disease Lamziekte in Cattle.—*Second Report of the Director of Veterinary Research. Union of South Africa.* Oct., 1912. pp. 79-160. (1913. Cape Town: Cape Times, Ltd., Government Printers.)

Leishmaniasis.

- (540) CANNATA (S.). Reperto del Parassita di Leishman nel Sangue Periferico. [The Discovery of Leishmania in the Peripheral Blood.]—*Pathologica*, 1913. June 15. No. 5. No. 111, pp. 351-352.
- (541) GABBI (U.). Au Sujet de l'Histoire du Kala-azar Méditerranéen.—*Bull. Soc. Path. Exot.*, 1913. March. Vol. 6. No. 3, pp. 141-143.
- (542) ——. On the Identity of Infantile and Donovan's Leishmania (Kala-Azar).—*Il. Trop. Med. & Hyg.*, 1913. July 1. Vol. 16. No. 13, pp. 198-199.
- (543) ——. Ueber den Ursprung der Leishmaniosis interna (Kala-azar) vom Hunde. [The Origin of Internal Leishmaniasis (Kala-azar) in the Dog.]—*Centralbl. f. Bakt.*, 1. Abt., Orig., 1913. July 3. Vol. 69. No. 7, pp. 504-516.

Rabies.

- (544) LUZZANI (L. N.). La Diagnosi della Rabbia con la Dimostrazione del Parassita specifico. [The Diagnosis of Rabies and the Demonstration of the Specific Parasite.]—*Pathologica*, 1913. May 1. Vol. 5. No. 108, pp. 253-259.
- (545) MIESSNER (H.). Immunisierungsversuche bei Tollwut. [Immunisation experiments against Rabies.]—*Berlin Tierärztl. Wochenschr.*, 1913. April 17. Vol. 29. No. 16, pp. 287-289.

Rabies—continued.

- (546) PREILER (W.). Neue Immunisierungsversuche bei Tollwut. [New Immunisation Experiments against Rabies.]—*Berlin Tierärztl. Wochenschr.*, 1913. April 3. Vol. 29. No. 14, pp. 249-252; and No. 15, pp. 269-273.
- (547) — & KAPFERBERGER (G.). Versuche zur Immunisierung von Hunden gegen Tollwut. [The Immunisation of Dogs against Rabies.]—*Zeitschr. f. Infektionskrankh., Parasit. Krankh. u. Hyg. d. Haust.*, 1913. June. Vol. 13. No. 6, pp. 307-316.
- (548) PIRONE (R.). I Corpi di Negri nella Rabbia. Nota II. [Negri Bodies in Rabies.]—*Pathologica*, 1913. April 1. Vol. 5. No. 106, pp. 191-196.

Rinderpest.

- (549) KNUTH (P.). Ueber das Auftreten und die Bekämpfung der Rinderpest in der Gegenwart. [The Occurrence of Rinderpest and the Methods at present in use for dealing with Outbreaks.]—*Zeitschr. f. Infektionskrankh., Parasit. Krankh., u. Hyg. d. Haust.*, 1913. May 5. Vol. 13. No. 5, pp. 273-293; and No. 6, pp. 356-369.

Spirochaetosis.

- (550) KLEINE (F. K.) & ECKARD (B.). Ueber die Lokalisation der Spirochäten in der Rückfallfieberzecke (*Ornithodoros moubata*). [The Localisation of Spirochaetes in the Relapsing Fever Tick.]—*Zeitschr. f. Hyg. u. Infektionskr.*, 1913. May 20. Vol. 74. No. 2, pp. 389-394.

Trypanasomiasis.

- (551) AOKI (K.) & KODAMA (H.). Beitrag zur Frage der Immunisierung mit abgetöteten Trypanosomen. [Immunisation with Dead Trypanosomes.]—*Zeitschr. f. Immunitätsforsch. und exp. Therap.*, 1. Teil., Orig., 1913. July 26. Vol. 18. No. 6, pp. 693-700.
- (552) BATTAGLIA (Mario). Granuloma ulcerante nel Prepuzio del Coniglio da Infezione Tripanosomica (*Trypanosoma dromedari* Tripoli). [Ulcerating Granuloma of the Prepuce of the Rabbit infected with *T. dromedari* Tripoli.]—*Atti. d. R. Accad. Medico-Chirurgica di Napoli*, 1913. No. 1. [3 pp.]
- (553) CARPANO (M.). *Trypanosoma* tipo "*Theileri*" nei Bovini della Colonia Eritrea. [A Trypanosome of the "*Theileri*" type in Cattle in Colonia Eritrea.]—*Clinica Veterinaria*, 1913. May 30. Vol. 36. No. 10, pp. 439-452. With 1 plate of 6 figures.
- (554) HECKENROTH (F.) & BLANCHARD (M.). Recherches sur les Propriétés du Sérum des Malades atteints de Trypanosomiase au Congo français.—*Bull. Soc. Path. Exot.*, 1913. June. Vol. 6. No. 6, pp. 444-447.
- (555) JOHNS (Foster M.). On the Adult Forms of *Trypanosoma americanum* in Naturally Infected Animals.—*Amer. Jl. Trop. Diseases & Preventive Med.*, 1913. July. Vol. 1. No. 1, pp. 49-59. With 1 coloured plate.
- (556) KOLLE (W.), HARTOCH (O.), ROTHERMUNDT (M.), & SCHÜRMANN (W.). Ueber neue Prinzipien und neue Präparate für die Therapie der Trypanosomeninfektionen. Chemotherapeutische Experimentalstudien. [New Principles and Preparations for the treatment of Trypanosome Infections. Chemotherapeutic Experimental Investigation.]—*Deut. Med. Wochenschr.*, 1913. May 1. Vol. 39. No. 18, pp. 825-828.
- (557) LAVERAN (A.) & MARULLAZ (M.). Au Sujet du *Trypanosoma talpae*.—*Compt. Rend. Soc. Biol.*, 1913. May 16. Vol. 74. No. 17, pp. 1007-1008. With 1 text-figure.

Trypanosomiasis—continued.

- (558) LAVERAN (A.) & ROUDSKY (D.). Le Galyl [Tetraoxydiphosphaminodiarsenobenzene] dans les Trypanosomiasés.—*Bull. Soc. Path. Exot.*, 1913. July. Vol. 6. No. 7, pp. 502-505.
- (559) MESNIL (F.). A propos du Pouvoir protecteur des Sérums des Malades du Sommeil.—*Bull. Soc. Path. Exot.*, 1913. June. Vol. 6. No. 6, pp. 447-451.
- (560) ROSENTHAL (F.). Untersuchungen über die Genese des Rezidivs bei der experimentellen Trypanosomeninfektion. [The Production of Relapse in Experimental Trypanosome Infections].—*Zeitschr. f. Hyg. u. Infektionskr.*, 1913. June 26. Vol. 74. No. 3, pp. 489-537.
- (561) SCHILLING (C.) & RONDONI (P.). Ueber Trypanosomen-Toxine und Immunität.—*Zeitschr. f. Immunitätsforschung. u. exp. Therap.*, 1. Teil., Orig., 1913. July 26. Vol. 18. No. 6, pp. 651-665.
- (562) SCHUBERG (A.) & BÖING (W.). Ueber den Weg der Infektion bei Trypanosomen- und Spirochätenkrankungen. [The Method of Infection in Trypanosome and Spirochaete Infections].—*Deut. Med. Wochenschr.*, 1913. May 8. Vol. 39. No. 19, pp. 877-879.
- (563) UHLENHUTH (Paul) & EMMERICH (Emil). Ueber das Verhalten des Kaninchenhodens bei experimenteller Trypanosomen- und Spirochäteninfektion. [The Behaviour of the Testes of the Rabbit in experimental Infections with Trypanosomes and Spirochaetes].—*Deut. Med. Wochenschr.*, 1913. April 3. Vol. 39. No. 14, pp. 642-644.

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- (564) VIGANO (L.). Le Termoprecipitine del Micrococco Melitense. [Thermo-precipitin of the *Micrococcus Melitensis*].—*Clinica Veterinaria*, 1913. May 30. Vol. 36. No. 10, pp. 453-456.

Biting Insects.

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- (566) STRICKLAND (C.). The Bionomics of the Rat-Flea.—*Brit. Med. J.*, 1913. May 31. p. 1160.
- (567) — & MERRIMAN (G.). Report on Rat-Fleas in Suffolk and North Essex.—*Parasitology*, 1913. April. Vol. 6. No. 1, pp. 2-18. With 3 charts.

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- (569) MITZMAIN (M. B.). The Biology of *Tubanus striatus* (Fabricius), The Horse-fly of the Philippines.—*Philippine J. of Science*. Section B. Tropical Medicine, 1913. June. Vol. 8. No. 3, pp. 197-221. With 7 plates.
- (570) NEWSTEAD (R.). A new Tsetse Fly from the Congo Free State (*G. severini*, sp. n.); and the Occurrence of *Glossina austeni* in German East Africa.—*Ann. Trop. Med. & Parasit.*, 1913. June 10. Vol. 7. No. 2, pp. 331-334. With 2 text-figures.
- (571) ROUBAUD (E.). Supplément à la Répartition et à la Variation Géographique des Glossines. [Supplement to the Distribution and Geographical Variation of the Glossinae].—*Bull. Soc. Path. Exot.*, 1913. May. Vol. 6. No. 5, pp. 347-350.

Biting Insects—continued.

Flies—continued.

- (572) SHIRCORE (J. O.). On Two Varieties of *Glossina morsitans* from Nyasaland.—*Bull. Entom. Research*, 1913. May. Vol. 4. No. 1, p. 89.

Mosquitoes.

- (573) LEGENDRE (J.). Note sur les *Stegomyias* du Tonkin.—*Bull. Soc. Path. Exot.*, 1913. July. Vol. 6. No. 7, pp. 511-513.
- (574) STRICKLAND (C.). Revised List of Malayan Anophelines.—*Indian Jl. Med. Res.*, 1913. July. Vol. 1. No. 1, pp. 203-205.
- (575) THEOBALD (F. V.). Second Report on the Mosquitoes of the Transvaal.—*Second Report of the Director of Veterinary Research. Union of South Africa*. Oct., 1912. pp. 315-342. With 2 plates. (1913. Cape Town: Cape Times, Ltd., Government Printers.)

Ticks.

- (576) BEDFORD (G. A. H.). A Tick new to South Africa [*Ornithodoros megnini* Dugès (1881)].—*Second Report of the Director of Veterinary Research. Union of South Africa*. Oct., 1912. pp. 343-344. With 1 plate. (1913. Cape Town: Printed by the Government Printers.)
- (577) BISHOPP (F. C.) & WOOD (H. P.). The Biology of some North American Ticks of the Genus *Dermacentor*. [*D. hunteri*, *D. albipictus*, *D. nigrolineatus*.]—*Parasitology*, 1913. July. Vol. 6. No. 2, pp. 153-186. With 3 plates and 1 map.
- (578) CUNLIFFE (N.). The Variability of *Rhipicephalus pulchellus* (Gerstäcker, 1873), together with its Geographical Distribution.—*Parasitology*, 1913. July. Vol. 6. No. 2, pp. 201-216. With 6 text-figures.
- (579) NUTTALL (G. H. F.). Note on Colouration in Ticks.—*Parasitology*, 1913. April. Vol. 6. No. 1, pp. 49-51. With 1 plate (coloured).
- (580) ———. Observations on the Biology of Ixodidae. Part I. [Dealing with *Ixodes putus*, *I. canisuga*, *I. hazagonus*, *I. ricinus*, *Haemaphysalis leachi*, *H. punctata*, *Hyalomma aegyptium*, *Rhipicephalus appendiculatus*.]—*Parasitology*, 1913. April. Vol. 6. No. 1, pp. 68-120. With 2 text-figures.
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- (582) ———. Parthenogenesis in Ticks (Preliminary Note).—*Parasitology*, 1913. July. Vol. 6. No. 2, pp. 139-140.
- (583) ———. *Rhipicephalus appendiculatus*: Variations in Size and Structure due to Nutrition.—*Parasitology*, 1913. July. Vol. 6. No. 2, pp. 195-203. With 4 text-figures.
- (584) ROBINSON (L. E.) & DAVIDSON (J.). The Anatomy of *Argas persicus* (Oken, 1818). Part I.—*Parasitology*, 1913. April. Vol. 6. No. 1, pp. 20-48. With 6 plates and 2 text-figures.
- (585) WARBURTON (C.). On Four new Species and Two new Varieties of the Ixodid Genus *Haemaphysalis*. [*H. aborensis*, *H. howletti*, *H. aciculifer*, *H. kinneari*, *H. cornigera* var., *anomala* n. var., *H. inermis* var., *aponommoides* n. var.].—*Parasitology*, 1913. July. Vol. 6. No. 2, pp. 121-130. With 8 text-figures.

Helminthiasis.

- (586) BLANC (G. R.) & HEDIN (H.). Distomes de l'Intestin du Chien à Montpellier.—*Compt. Rend. Soc. Biol.*, 1913. May 2. Vol. 74. No. 15, pp. 884-885.
- (587) BRAU (P.). De l'*Anquillula intestinalis* en Cochinchine et de son Diagnostic Hématologique.—*Bull. Soc. Path. Exot.*, 1913. April. Vol. 6. No. 4, pp. 262-264.
- (588) BRUMPT (E.) & CAUCURTE (R.). Essais de Traitements préventifs des Strongyloses des Ruminants.—*Bull. Soc. Nationale d'Acclimatation de France*, 1912. June. [8 pp.]
- (589) GEDOELST (L.). Un Type nouveau de *Dicrocoeliid* parasite des Primates.—*Bull. Soc. Path. Exot.*, 1913. April. Vol. 6. No. 4, pp. 256-259. With 1 plate.
- (590) LÉGER (A.). *Gastrodiscus polymastos* Leuck., 1880 chez les Equidés du Haut-Sénégal et Niger.—*Bull. Soc. Path. Exot.*, 1913. April. Vol. 6. No. 4, pp. 261-262.
- (591) ———. Microfilaires sanguicoles de quelques Oiseaux du Haut-Sénégal et Niger.—*Bull. Soc. Path. Exot.*, 1913. May. Vol. 6. No. 5, pp. 359-367.
- (592) RAILLIET (A.) & HENRY (M.). Sur les Douves de l'Intestin du Chien.—*Compt. Rend. Soc. Biol.*, 1913. May 9. Vol. 74. No. 16, pp. 929-930.
- (593) ——— & ———. Sur les Oesophagostomiens des Ruminants.—*Bull. Soc. Path. Exot.*, 1913. July. Vol. 6. No. 7, pp. 506-511. With 4 text-figures.
- (594) ———, ———, & JOYEUX (C.). Un nouveau Strongylid des Singes.—*Bull. Soc. Path. Exot.*, 1913. April. Vol. 6. No. 4, pp. 264-267.
- (595) SKRJABIN (K. I.). *Schistosomum turkestanicum*, nov. sp. ein neuer Parasit des Rindes aus Russisch-Turkestan. [*Schistosomum turkestanicum*, n. sp. of Cattle in Russian Turkestan.] —*Zeitschr. f. Infektionskrankh. Parasit. Krankh. u. Hyg. d. Haust.*, 1913. July. Vol. 13. No. 7, pp. 457-468. With 2 plates.

Protozoa.

- (596) BRUMPT (E.). A propos de l'*Haemocystozoon brasiliense* de Franchini.—*Bull. Soc. Path. Exot.*, 1913. June. Vol. 6. No. 6, pp. 377-380. With 17 text-figures.
- (597) BALFOUR (Andrew). A Sarcocyst of a Gazelle (*G. rufifrons*) showing Differentiation of Spores by Vital Staining.—*Parasitology*, 1913. April. Vol. 6. No. 1, pp. 52-56. With 2 plates.
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- (599) FANTHAM (H. B.). Note on the Specific Name of the Herpetomonas found in the Dog-Flea, *Otenocephalus canis*.—*Bull. Soc. Path. Exot.*, 1913. April. Vol. 6. No. 4, pp. 254-255.
- (600) LÉGER (M.). Hématozoaires d'Oiseaux de la Corse.—*Bull. Soc. Path. Exot.*, 1913. July. Vol. 6. No. 7, pp. 515-523.
- (601) MARTOGLIO (F.). Contributo alla Conoscenza delle Leucocitogregarine.—*Ann. d'Igiene Speriment.*, 1913. Vol. 23. No. 2, pp. 161-170. With 1 coloured plate.
- (602) PHISALIX (M.). Sur une Hémogrégarine du Python molure et ses Formes de Multiplication endogène.—*Compt. Rend. Soc. Biol.*, 1913. May 23. Vol. 74. No. 18, pp. 1052-1054. With 15 text-figures.

Protozoa—continued.

- (603) PHISALIX (M.). Sur une Hémogrégarine de la Vipère Fer de Lance et ses Formes de Multiplication endogène.—*Compt. Rend. Soc. Biol.*, 1913. June 20. Vol. 74. No. 22, pp. 1286-1288. With 1 text-figure.
- (604) — & LAVERAN (A.). Sur une Hémogrégarine nouvelle de *Lachesis alternatus*.—*Bull. Soc. Path. Exot.*, 1913. May. Vol. 6. No. 5, pp. 330-333. With 12 text-figures.
- (605) RONDONI (P.). Sulla Classificazione dei Protozoi Emoparassiti. Il nuovo ordine dei *Binuclati* (Hartmann). [The Classification of the Protozoal Parasites of the Blood. The New Order *Binuclata* (Hartmann).]—*Lo Sperimentale*, 1913. April 7. Vol. 67. No. 1, pp. 105-118.
- (606) THIROUX (A.). Les Formes de Reproduction par Schizogonie et Sporogonie d'*Haemogregarina pectiti* (Thiroux, 1910) chez *Crocodilus niloticus*.—*Bull. Soc. Path. Exot.*, 1913. May. Vol. 6. No. 5, pp. 327-330. With 10 text-figures.
- (607) VIGUIER (G.) & WEBER (A.). Les Mitochondries de l'*Haemogregarina sergentium* durant son Evolution dans le Sang du Gongyle.—*Compt. Rend. Soc. Biol.*, 1913. April 11. Vol. 74. No. 12, pp. 664-666.
- (608) — & —. Nouvelles Observations sur l'Altération des Hématies sous l'Influence d'une Hémogrégarine chez le Gongyle.—*Compt. Rend. Soc. Biol.*, 1913. April 18. Vol. 74. No. 13, pp. 760-761.

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- (609) ANDREWS (W. H.). Experiments with Snakes.—*Second Report of the Director of Veterinary Research. Union of South Africa*. Oct., 1912. pp. 406-483. With 4 plates. (1913. Cape Town: Cape Times, Ltd., Government Printers.)
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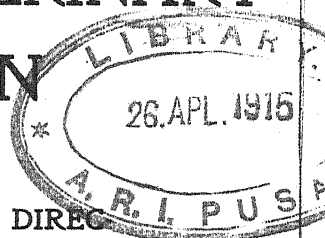
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ERRATA.

No. 4, p. 181, lines 3 and 4: the word "resistant" should precede the word "and."

p. 183, para. 3, line 5: for 1-10 cc. read 1·1 cc.

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[No. 1.

BABESIASIS (PIROPLASMOSIS) AND ANAPLASMOSIS.

- (1) SERGENT (Ed.), LHÉRITIER (A.), & ISMERT (R.). *Études sur les Piroplasmoses en Algérie. 1. Sur la Piroplasmose équine en Algérie. Guérison par le Trypanbleu.* [The Piroplasmoses in Algeria. 1. Equine Piroplasmosis. Recovery after Treatment with Trypanblue.]—*Bull. Soc. Path. Exot.* 1913. Oct. Vol. 6. No. 8. pp. 571-572.

This disease was first recognized in Algeria by ROGER in 1906. In 1913 the authors found a case on a farm near Algiers in a 14-year-old horse which had not left the premises for seven years.

The first symptom, consisting of oedema of dependent parts, was first seen on June 15th, and an examination of the blood six days later showed that 4 per cent. of the blood corpuscles were invaded by piroplasms. A subcutaneous injection of 5 grammes of trypanblue was given on the same day.

On the following day the animal was in about the same condition and an examination of the blood shewed that about 1 per cent. of the corpuscles contained parasites. A second dose of 5 grammes of trypanblue was administered intravenously.

Two days later there was considerable improvement in the animal's condition. The number of invaded corpuscles had fallen to about 0.3 per cent., and many of the parasites appeared to be degenerating.

By June 25th parasites seemed to have entirely disappeared from the peripheral blood, and recovery was complete a few days later.

Bovine piroplasmosis is known to exist in the district, but no case of the equine disease has been recorded previously.

- (2) SERGENT (Ed.) & BEGUET (M.). *Études sur les Piroplasmoses d'Algérie. 2. Existence d'*Anaplasma marginale* Theiler chez les Boeufs d'Algérie.* [The Existence of Anaplasmosis in Cattle in Algeria.]—*Bull. Soc. Path. Exot.* 1913. Oct. Vol. 6. No. 8. pp. 573-574.

The authors have examined a large number of blood preparations made from cattle shewing symptoms of piroplasmosis, obtained from a number of different districts, and have discovered anaplasma in three of them.

In one instance the blood of an animal shewing symptoms of piroplasmosis was found to contain anaplasma, 8 per cent. of the corpuscles being invaded. No piroplasms were found.

In the second case five animals were found to be infected, the percentage of infested corpuscles varying from 4 to 70. In this case also no piroplasms were found.

In the third instance the number of invaded corpuscles was 15 per cent.

In every instance there was polychromatophilia, metachromasia, and anisocytosis.

- (3) SERGENT (Edm.), LHÉRITIER (A.), & BOQUET (A.). *Études sur les Piroplasmoses en Algérie. 3. Essais de Traitement de la Piroplasmose Bovine par le Trypanbleu.* [The Treatment of Bovine Piroplasmosis by Trypanblue.]—*Bull. Soc. Path. Exot.* 1913. Nov. Vol. 6. No. 9. pp. 618–622.

The authors state that, while the symptoms of Algerian bovine piroplasmosis will be discussed in a future paper, certain cases have been encountered which present a typical picture of the disease from a clinical standpoint, but in which piroplasms are either very scantily present in the blood, or quite absent. The diagnosis is justified by the clinical features presented and by the presence of other cases in which the parasite can be discovered. Further evidence is afforded by the exclusion of other diseases.

The cases described are divided into two main classes, viz., those in which the injection of the dye has appeared to be of some value, and those in which it has been valueless.

The first group contains mild or moderately severe cases, and the latter the severe cases. Of these there are three types, those in which piroplasms are very numerous in the blood, cases in which they are scanty, and cases in which no parasites can be found.

No description of the parasite is given in this paper, but mention is frequently made of small ring and bacillary forms, and, further, in one instance it is said that 95 per cent. of the red corpuscles were invaded. From this it appears to be not improbable that the author was dealing with a disease of the East Coast Fever type. Haemoglobinuria is said to have occurred in one case only, but it is not specified in what form of the disease this occurred.

Even in cases in which the injection of the drug has appeared to be beneficial the temperature did not fall for several days after. No alteration in the morphology of the parasite has been observed as a result of the injection of the dye.

- (4) SERGENT (Edm.), & LHÉRITIER (A.). *Études sur les Piroplasmoses en Algérie. 4. Infection piroplasmique intense chez des Bovidés ne présentant aucun Symptôme morbide.* [Heavy Infection with Piroplasms in Cattle shewing no Symptoms of Disease.]—*Bull. Soc. Path. Exot.* 1913. Nov. Vol. 6. No. 9. pp. 622–623.

The authors have made frequent examinations of the blood of a number of Breton cattle during a period of several months dating from their arrival in Algeria.

At the time of their arrival examination of the blood shewed no parasites, but during the summer following parasites were found to be present in the blood, in some cases in large numbers.

A table is given shewing the percentage of infected corpuscles found in nine animals which were examined about once a month during the year. In six instances during the months December 1911 and April 1912 parasites were found in 0·05 per cent. of the blood corpuscles, but in July 1912 the number varied from 1 per cent. to 80 per cent. During this month eight of the animals were affected, and the ninth had shewn 7 per cent. of infected corpuscles during June. Towards the end of July the number had fallen again. According to the record given only two animals were examined on this occasion, and 2 per cent. of invaded corpuscles were found in one of them.

The only description given of the piroplasms is that they were small, and round or bacilliform.

- (5) CHAMBERS (F.). **Immunisation of imported Cattle against Northern Rhodesia Piroplasmosis and Anaplasmosis.**—*Jl. Comp. Path. & Therap.* 1913. Sept. Vol. 26. No. 3. pp. 249–253.

After noting the results obtained by THEILER and BEVAN the author gives the history of two imported bulls which he inoculated against the two diseases. It was the author's intention to inoculate them with a strain of anaplasma (*centrale* variety) obtained from Pretoria, but owing to loss in the post he was unable to do so. The animals on arrival were quarantined and were kept as free as possible of ticks; only eight of the *decoloratus* variety being found on them. On the 18th day they were inoculated subcutaneously with 25 cc. of citrated blood taken from an ox brought down from Barotseland a short time previously. Both animals contracted babesiasis (redwater) and one was given an injection of trypanblue. The subsequent anaplasmosis reaction was more severe in this animal than in the other. The anaplasmosis reaction caused considerable constitutional disturbance in both animals, but treatment with quinine and iron was resorted to and both made good recoveries. They were forwarded to the owner on the 34th day after inoculation.

- (6) TRAUTMANN (O.). **Anaplasmosis der Schafe in Deutsch-Östafrika.** [Anaplasmosis of the Sheep in German East Africa.]—*Berlin. Tierärztl. Wochenschr.* 1913. Aug. 14. Vol. 29. No. 33. pp. 593–594.

The author claims to be able to confirm SCHELLHASE's observations regarding the occurrence of this disease, and states that he observed it two years ago, though he was at the time unable to carry his investigations through.

He states that the disease has a wide distribution in Kondoa-Irangi. Young animals are principally attacked. The symptoms are:—A greenish-yellow discharge from the eyes and nose, progressive wasting, and in some cases diarrhoea. As the wasting progresses there is observed oedema of dependent parts. The disease sets in during very dry seasons and tends to disappear with the onset of the rains.

Examination of the blood shews the presence of a marginal anaplasma with a morphology exactly resembling that described by THEILER and SIEBER.

The usual abnormalities are to be observed in the blood and the lesions are those of an acute anaemia.

- (7) DESCAZEUX (M. J.). **Considerations étiogéniques, pathogéniques et thérapeutiques sur la Piroplasmose des Bovidés dans l'Etat de Saint Paul.**—*Recueil de Méd. Vét.* 1913. Nov. 30. Vol. 90. No. 22. pp. 392-410.

Piroplasmosis in bovines in Saint Paul is invariably due to *Piroplasma* (*Babesia*) *bigeminum*, although CARINI has described the occurrence of *P. argentinum* in an imported animal.

The disease occurs in all parts of the country, and has been responsible in some instances for a mortality of 95 per cent. in imported animals. Adult animals are more susceptible than young ones. The transmitting tick is *Margaropus annulatus* var. *australis*. The author gives a detailed description of both sexes of this parasite.

Piroplasms are to be found in experimental cases at the seat of inoculation from the third or fourth day, although they do not appear in the circulating blood until the 7th or 8th day. These parasites are small and rounded and are often extra-corpuseular.

It is said that in cases of natural infection piroplasms can be found in blood taken from the ears before they appear in the general circulation, the explanation being that the infecting ticks are commonly found on the ears and the parasites found there are those injected by the tick before the circulation becomes invaded. It is also stated that this explains why intravenous injection of trypanblue is more effective than subcutaneous injection in natural cases of the disease.

In the treatment of the disease with this drug the author has had four deaths out of 15 natural cases and none out of 60 experimental infections. The drug is said to be more efficient when it is injected close to the seat of inoculation.

Auto-agglutination of the red corpuscles occurs before piroplasms can be detected in the blood, and becomes very marked when piroplasms are numerous.

The effect of trypanblue is to act directly upon the parasites and to cause very marked phagocytosis.

It is said that this phagocytosis is observed in animals that have been immunised against piroplasmosis, treated with trypanblue and immediately infected with anaplasma.

Parasites disappear from the blood about 20 hours after the injection of trypanblue, but they always reappear as small round bodies during the 10 to 20 days following recovery.

- (8) CARPANO (M.). **Piroplasmose equina, Typi parassitari.** [Equine Piroplasmosis. The Types of the Parasites.]—*La Clinica Veterinaria.* 1913. Oct. 15/30. Vol. 36. No. 19/20. pp. 847-886. With 3 plates.

The author records the occurrence in Italy of the two types of parasites that are responsible for this disease and which have been encountered in other parts of the world. He has been able to satisfy

himself that they are distinct since he has found that animals immune to one are susceptible to infection with the other. A large proportion of native Italian horses appear to be immune to piroplasmosis, possibly as a result of infection while foals.

Clinically there does not appear to be much difference between the two types of infection, save that *Babesia caballi* appears to be responsible for more chronic cases than *Nuttallia equi*, which is the cause of more deaths. The immunity lasts for a considerable time.

The transmitting agent is *Rhipicephalus bursa*, and *Boophilus annulatus* appears to be capable of transmitting *Babesia* only.

- (9) DARLING (S. T.). **Equine Piroplasmosis in Panama.**—*Jl. Infectious Diseases*. 1913. Sept. Vol. 13. No. 2. pp. 197–202. With 1 plate.

In this paper the author describes the occurrence of a case of equine piroplasmosis at Ancon.

The animal in question was one of the first to be sent to the Canal Zone in 1904. In January 1913 it was taken to Old Panama and the Juan Diaz River, and while there was allowed to graze on the Sabannas where there were some native ponies. Four days later the animal was found to be ill with an elevated temperature. The pulse and respiration were accelerated, and after an illness lasting three days the horse died.

At the post-mortem the following lesions were found :—

No subcutaneous oedema or fluid in the serous cavities. The anterior surface of the liver shewed a number of purplish areas of necrosis measuring up to rather more than an inch in diameter and with yellow centres. None were found in the substance of the liver. The spleen was enormously enlarged, and its pulp was dark in colour and rather fluid. Its weight was estimated at 28 pounds. The blood was somewhat more fluid than normal, and on microscopic examination there appeared to be some amount of leucocytosis, with a predominance of transitional and mononuclear forms. The parasite found was that described by NUTTALL and STRICKLAND as *Piroplasma (Babesia) caballi*; parasites were scanty in the blood. Ticks removed from the horse at the time of death were identified by NUTTALL and WARBURTON as *Dermacentor nitens* and *Amblyomma cajannense*.

- (10) LAVERAN (A.) & NATTAN-LARRIER. **Piroplasmoses Canines d'Europe et d'Afrique.** [The Canine Piroplasmoses of Europe and Africa.]—*Ann. Inst. Pasteur*. 1913. Sept. Vol. 27. No. 9. pp. 701–717.

This paper contains a description of the experiments undertaken with the object of deciding whether the European and African canine piroplasmoses are identical or not.

The strains used were a French strain and a strain from Cape Colony. In order to attenuate the virulence of the piroplasms somewhat, the blood used for inoculation was placed in an ice chest for from three to seven days before it was used.

French virus. The period of incubation varied from two to five days, and of 23 dogs inoculated eight failed to become infected.

Two forms of the disease are distinguished, an acute and a sub-acute.

The acute form was most frequently seen, particularly after the second passage. Piroplasms appeared in the blood in from two to four days and death took place in from one to four days later.

In the subacute form of the disease crises occurred in from four to seven days after the appearance of parasites in the blood, and in some cases there were relapses at varying intervals afterwards. As a rule the parasites were very scanty during the relapses. Jaundice was observed in two cases only, but Gmelin's reaction was obtained with the urine containing haemoglobin in every instance.

Haemoglobinuria occurred in every dog that contracted the disease fatally, while in no case was it observed where the animal recovered.

In the acute form there are three constant lesions: anaemia, enlargement of the spleen, and congestion of the kidneys.

African virus. This virus, which was obtained from NUTTALL, was derived from a chronic case of the disease. As in the case of the French virus the inoculations were all intramuscular.

During the first five passages the period of incubation varied from 6 to 14 days, but subsequently it was reduced to 4 or 5, reaching 7 days in one instance only, when the blood had been kept in the ice chest for more than a week.

Excluding two cases in which the course of the disease was subacute, the average duration of the disease was 12 days, death occurring on the third day after the appearance of parasites in the blood.

Two dogs failed to become infected.

The acute form of the disease was most frequently observed, this type of infection occurring in two-thirds of the cases subsequently to the fourth passage.

The disease ran a sub-acute course in three cases only, and in two of these was followed by recovery. In these two cases the dogs were fairly old and one of them failed to become infected by the first inoculation. The second of these animals was inoculated with blood that had been preserved on ice for 10 days.

In almost every case in which the animals suffered from an acute attack of the disease there was observed haemoglobinuria, but jaundice was noted in one instance only. Bile was detected in the urine in 7 cases.

The lesions found were the same as those observed in the animals infected with the French virus.

Examination of the records shews that the only difference to be observed between the results of inoculation with the two viruses is that the African virus appears to be the more virulent. Upon these grounds cross-immunity experiments were undertaken. None of the dogs which had recovered either from the French or the African disease became infected when re-inoculated with the same strain. A bitch which had survived inoculation with both strains whelped, and one of the puppies inoculated immediately after birth with the French strain failed to become infected, while another inoculated 13 days after birth with the African strain contracted a fatal infection. The authors conclude from this that there is an immunity immediately after birth which soon disappears. In two instances the authors have shewn that the piroplasm does not pass from the mother to the foetus.

In the cross-immunity experiments dogs which had resisted infection with either strain were re-inoculated with the same strain 10 days

later, and then again inoculated with the other strain after a lapse of a further period of about ten days.

Details of these inoculations are given, and the results shew that of 7 dogs which had acquired immunity to the French strain 6 died of the disease when inoculated with the African strain, and 2 which had acquired immunity to the African strain suffered from a slight attack when inoculated with the French strain.

The authors conclude that these experiments shew a marked difference between the two strains. They point out that, if the difference was one of degree of virulence only, the immunity conferred by recovery from the disease caused by the French strain would cause the attack produced by the African strain to be of a mild character, whereas the contrary proved to be the case. Further, recovery from the African disease should confer absolute immunity against the French form of disease. This was also found not to be the case.

They conclude that the African parasite, if not a distinct species, is at least a variety distinct from the French parasite.

- (11) KNUTH (P.) & RICHTERS (E.). Ueber die Vermehrung von *Piroplasma canis* auf künstlichen Nährböden. [The Cultivation of *Piroplasma* (*Babesia*) *canis* in Artificial Culture Media.]—*Zeitschr. f. Infektionskr. parasit. Krankh. u. Hyg. d. Haust.* 1913. Sept. Vol. 14. No. 2/3. pp. 136-144. With 2 plates.

While passing under rapid review the literature regarding the cultivation of malarial parasites and piroplasms, the authors state that their own experiments were commenced in February 1913 and were carried out independently of ZIEMANN.

Owing to differences in the characters possessed by piroplasms as compared with those of malarial parasites of man the authors have not closely adhered to the technique described by BASS and JOHNS. They point out that while piroplasms can under certain circumstances retain their vitality outside the body for a long time, the malarial parasites rapidly die, and also that the processes of multiplication of the two types of organism in the blood are very different.

It is laid down that in order to get successful cultures it is essential that there shall be only a small number of parasites present in the blood used, for if piroplasms are numerous the vitality of the organism is lowered and large quantities of inhibiting substances are present in the blood.

In order to get suitable blood it is advised that a young dog, from four to eight weeks old, should be inoculated, and if only old dogs are available for experiment, splenectomy should first be performed, and after a day or two 40 to 50 cc. of blood should be withdrawn in order to produce a temporary anaemia.

Repeated estimations have not shewn that piroplasms are more numerous in arterial blood than in venous blood, or vice versa. Blood is withdrawn from the jugular vein and mixed with half its volume of 1 per cent. dextrose solution. The mixture is defibrinated and centrifuged, the serum and leucocytes being pipetted off. The tubes are then incubated at 40° to 41° C. Multiplication occurs during the first eight hours, and numerous, pear-shaped, elongated, and amoeboid intracorpuseular forms are to be found. Extracorpuseular forms

may also be encountered. Both the corpuscles and the parasites begin to degenerate by the 20th to 24th hour.

Subsequently certain alterations were made in the technique. The serum was not removed, a 2 or 3 per cent. solution of dextrose was used, and 1/10 cc. of 2 per cent. citrate solution was added to 5 cc. of blood. The best results were obtained when the tubes were left at room temperature, multiplication being observed up to the 6th or 8th day.

Numerous other media were tried, and ascetic fluid mixed with serum inactivated at 45° C. for an hour (proportions not stated) was found to be a good medium.

Subsequently the proportion of dextrose was increased, 1/10 cc. of a 50 per cent. solution being added to 6 cc. of blood.

Subcultures were obtained in media of composition similar to those used for the primary cultures, and it was found that the best results were obtained when the secondary cultures were made on the second or third day.

It was found possible to inoculate dogs successfully with sub-cultures and also to cultivate the piroplasms from dogs so infected.

(12) TOYODA (H.). *Züchtungsversuche mit Babesia canis nach der Bassschen Methode.* [The Cultivation of *Babesia canis* by Bass's Method.]—*Centralb. f. Bakt.* 1. Abt., Orig. 1913. Nov. 26. Vol. 72. No. 1/2. pp. 76–81. With 1 plate.

The author has succeeded in obtaining cultures in the following manner:—

Tubes were prepared containing the following mixture: 10 cc. of serum from a healthy dog, 0.2 cc. of 50 per cent. dextrose solution, 0.3 cc. of 2 per cent. sodium citrate in 0.85 per cent. sodium chloride. These tubes were inactivated by incubation at 45° C. for an hour.

The blood used as seed material was treated in the following manner. Blood in which parasites had just made their appearance was withdrawn from a vein and defibrinated, and then mixed with the same materials as the healthy serum and in the same proportions. The tubes were centrifuged, and after removal of the serum and leucocytes the corpuscles were added in the proportion of 0.4 to 0.5 cc. to the tubes of serum previously prepared.

In the subcultures 0.4 cc. of freshly prepared corpuscles from a healthy dog was mixed with 0.1 cc. of the "culture-blood," the mixture being incubated at 37° C.

Details are given of a particular case from which the following particulars are taken:—

The seed material (infected blood) which was used was examined microscopically, and in 50 fields (magnification not given) 6 parasites were found. In other preparations of the same blood several parasites which had divided into 2, 4, and 8 were found. The cultures were maintained for the first 18 hours at 37° C. and then for 6 hours at room temperature, this alternation of temperature being continued.

After 18 hours parasites were found at four places in 50 fields and they shewed from 6 to 8 merozoites, the total number observed being 25.

On the second day 46 merozoites were found in 50 fields, some of them being in groups of 16. The parasites shewed an increase in size, and in many instances one end was drawn out into a fine point.

On the third day the majority of the parasites were extracorpuscular and degenerated, but at one place 20 merozoites in a condition of partial degeneration were found in a single corpuscle.

No parasites were to be found on the 6th day.

The first subculture was made by inoculating fresh tubes from the primary culture at the 18th hour. It is not stated whether the alternation of temperature was used, but multiplication occurred as shewn by the examination of films. The multiplication was not so marked as in the primary culture and by the 4th day the majority of the parasites were degenerated.

The second subculture was made from the first at the 18th hour. After 18 hours incubation there appeared to be no multiplication, but after 48 hours at room temperature there was obvious multiplication. The cultures were then incubated at 37° C. for 48 hours and further multiplication occurred. Young schizogonous forms comprising only 2 or 4 merozoites were found, indicating that fresh blood corpuscles had been invaded.

Degeneration of the parasites occurred by the 5th day. No multiplication occurred in the third subculture.

THEILERIASIS.

- (13) NUTTALL (G. H. F.) & HINDLE (E.). **Conditions influencing the Transmission of East Coast Fever.**—*Parasitology*. 1913. Oct. Vol. 6. No. 3. pp. 321-332.

The authors have made an attempt to confirm GONDER's account of the life cycle of *Theileria parva* within the invertebrate host, but they have been unable to detect in the tissues of infected ticks any appearances which could not be found in the corresponding tissues of clean ticks. More than 20 ticks have been submitted to thorough examination.

In order to throw more light upon the biology of the parasite in the tick, a number of experiments were undertaken to ascertain the effect of various conditions upon the infectivity of the ticks. Full protocols of the experiments are given and these afford the following information.

In the first experiments it was shewn that ticks do not necessarily become clean as a result of biting a vertebrate host, and further that an infected tick does not become infective until it has fed upon a bovine for at least two days.

Secondly it was shewn that infected ticks could be fed upon a rabbit for three days before being placed upon a susceptible animal, without losing their power of infecting the bovine. In the experiments given the period of incubation was exactly the same as that in the control animal, excluding the three days spent by the ticks upon the rabbit.

Infected ticks were placed upon a susceptible calf and were removed after they had been attached for two days. The calf did not become infected, but was proved subsequently to be susceptible to infection.

It being thought possible that the non-infectivity of the ticks during the first two days spent upon a bovine animal might in some way be connected with the temperature, unfed infective ticks were heated to 37° C. for two days before they were placed upon a susceptible animal,

but this did not render them infective during their first two days spent upon it.

The fifth experiment shewed that preliminary feeding of infective ticks for two days, followed by starvation for 17 days, rendered them non-infective.

Since experiments shewed that ticks are non-infective until after they have fed for two days, it appeared to be reasonable to suppose that the failure of attempts to infect cattle by the inoculation of ticks was owing to the inoculation of parasites at an unsuitable stage of development.

Two calves were inoculated respectively with the contents of 19 engorged ticks and the salivary glands of 18 infected ticks. Neither of these became infected, but the calf upon which they were fed became infected and died.

Some experiments were carried out to test the effect of cold upon the infectivity of *R. appendiculatus* infected with *T. parva*.

Engorged larvae after being kept for about five weeks at 19° C. were found to be infective after being warmed for a single night to 37° C.

Infected ticks were found to be non-infective after having been maintained at about 10° C. for three weeks. It was found that warming the ticks thus rendered non-infective restored their infectivity.

TRYPANOSOMIASIS.

- (14) BRUCE (David), HARVEY (D.), HAMERTON (A. E.), & Lady BRUCE. **Trypanosomes of the Domestic animals in Nyasaland. I. *Trypanosoma simiae*, sp. nov. Part II.—The Susceptibility of Various Animals to *T. simiae*.—*Proc. Roy. Soc.* 1913. Oct. 1. Series B. Vol. 87. No. B. 592. pp. 48-57.**

The morphology of this parasite has been dealt with in a previous paper (see this *Bulletin* Vol. 1. No. 2. pp. 77-78); the present publication deals with the pathogenic range of the trypanosome.

The organism kills monkeys and the domestic pig in an extremely short time, but is non-pathogenic for cattle, antelope, dogs, and small experimental animals. The trypanosome is however capable of infecting the goat and the sheep.

So far as is known, the warthog is the only wild animal in the district which harbours it, and this animal is very numerous present in the low country. This explains why so large a number of infected flies are found.

The trypanosome undergoes a change of virulence with great rapidity. If a monkey and a goat be infected by means of flies, the monkey dies within a few days, but if an attempt be made to infect a monkey by inoculation with the goat's blood, the result is generally a failure.

"The organism belongs to the same group as *T. pecorum*, and it is curious that this loss of virulence occurs in the latter species also. . . . This has given rise to the erroneous idea that a separate species—*T. nanum*—exists. *T. nanum* is in truth nothing but a strain of *T. pecorum* which has lost its virulence for these animals (monkey, dog, rat) by its passage through the goat."

Tables are given shewing the results of a large number of experimental inoculations.

The following is an abstract of the summary of the results obtained :—

The opportunity has not yet been offered to test the effect of *T. simiae* upon equines. Four oxen have been inoculated, two from a monkey and two from a goat, and all have remained in good health. Five antelope have been inoculated without positive results, and the trypanosome has never been found in the blood of an antelope.

Thirty-one goats and one sheep have been infected. Passage through the fly would appear to exalt the virulence, or at least the trypanosome would appear to reach its highest virulence after passage through the fly. Of 15 goats infected by flies, 13 died in, on an average, 46 days. Three goats inoculated directly from warthogs all recovered; this is curious, since in all probability the fly must get its infection from that species. Six goats were inoculated with blood from fly-infected monkeys. One died, four recovered, and one was refractory. Three goats out of six infected from goats died, the remainder recovering.

Of the cases of fly infection in the goat 12 were cases of mixed infection, but from the results obtained it would appear that *T. simiae* alone is as fatal as when mixed with other trypanosomes. The only symptoms presented by goats are emaciation, anaemia, and exhaustion.

In the domestic pig the disease runs a very rapid course. In nine experiments the average period elapsing between inoculation and death was 5·3 days.

When monkeys are infected directly from flies death takes place within a fortnight, and a similar result is obtained when monkeys are inoculated from other monkeys which have very recently been infected by the bite of flies. When a monkey is inoculated with the blood of an infected goat the result is generally negative.

The dog appears to be immune, as do the guinea-pig and rat. The rabbit presents a high degree of resistance.

The carrier is *G. morsitans*; 3·4 per 1,000 were found to be infected. The reservoir of the virus appears to be the warthog. Of 33 examined three were infected.

- (15) BRUCE (David), HARVEY (D.), HAMERTON (A. E.), & Lady BRUCE. **Trypanosome Diseases of Domestic Animals in Nyasaland. I. *Trypanosoma simiae*, sp. nov. Part III.**—*Proc. Roy. Soc.* 1913. Oct. 1. Series B. Vol. 87. No. B. 592. pp. 58–66. With 3 plates.

In this paper the development of *T. simiae* is described. The trypanosome belongs to the pecorum group. In this group development takes place in the intestinal tract, including the labial cavity of the proboscis, and afterwards the trypanosomes reach the hypopharynx, where they revert to the original blood or infective form.

Flies were found to become infective in 50 days at a temperature of 62° F. and in 20 days at 83° F.

By causing flies to salivate upon coverglasses the long narrow intestinal form of the organism was found, and there is no doubt that the fly has the power of regurgitating the contents of the proventriculus and intestine forwards into the labial cavity and probably into the blood stream.

Conclusions :—

- “ 1. That *T. simiae* can be transmitted from infected to healthy animals by the tsetse fly *G. morsitans*.
- “ 2. That *T. simiae* multiplies in the intestines and in the labial cavity of the proboscis of the fly. Here only developmental forms are found, never infective forms.
- “ 3. That *T. simiae* growing in the intestines of the “ fly ” has no specific characters by which it can be distinguished from other species of pathogenic trypanosomes found in tsetse flies.
- “ 4. That the final stage of the development takes place in the hypopharynx, wherein the infective form of the parasite, similar in shape to the trypanosome found in the blood of infected animals, is produced.
- “ 5. That the flies do not become infected until about 20 days after their first infected feed.”

- (16) BRUCE (David), HARVEY (D.), HAMERTON (A. E.), & Lady BRUCE. **Trypanosome Diseases of Domestic Animals in Nyasaland. III. *Trypanosoma pecorum*.**—*Proc. Roy. Soc.* 1913. Oct. 1. Series B. Vol. 87. No. B. 592. pp. 1–26.

This trypanosome has been found in cattle, wild game, and wild flies in the neighbourhood of Kasu, and it is one of the most important trypanosomes of domestic animals in Central Africa, since it affects horses, cattle, goats, sheep, pigs, and dogs. The morphology of the trypanosome has already been dealt with. The bulk of this paper is made up of tables of measurement of the trypanosome in various animals and charts constructed upon these. A comparison of the charts constructed upon the measurements of the parasite occurring in domestic cattle, wild game and wild *G. morsitans* shews that they are very similar. The measurements were taken from the parasites present in the blood of a number of different animals.

The cattle strain of the organism is fatal to goats in 30 to 40 days, but death does not appear to be the invariable result.

Dogs are certainly killed within 30 days. Guinea-pigs and white rats appear to offer an absolute resistance to infection.

The wild game strain produces the same effect upon the goat as the domestic cattle strain. The monkey offers a considerable resistance. Only three out of eleven monkeys died of the infection, the remainder never shewing trypanosomes at all. In these three animals the period of incubation varied from 9 to 27 days, and the duration of the disease from 88 to 236 days.

Nine goats and nine dogs were inoculated with the wild game strain, and while all the goats became infected, none of the dogs contracted the disease. Of nine rats inoculated with this strain four remained healthy and five died in about 30 days.

Experiments with the wild fly strain had the following results: A single ox sent into the “ fly ” country contracted a double infection of *T. pecorum* and *T. caprae*, and died in 140 days.

Flies brought up from the “ low-country ” were as a rule fed upon a goat, a monkey, and a dog. As the flies were often infected with several species of trypanosome, the result in many cases was one of mixed infection. In 50 experiments with goats, 10 were pure infections with *T. pecorum*. As *T. pecorum* is the most rapidly fatal trypanosome for the goat among those occurring in the district, it may be assumed that it was mainly responsible for death in the 28 cases in which the infection was mixed.

In eight feeding experiments with wild *G. morsitans* carried out with pigs, only two shewed a pure *pecorum* infection.

Only six monkeys out of 28 became infected with this strain.

The dog is about as susceptible as the goat, and death usually takes place in, on an average, 41 days.

The carrier of the infection is *G. morsitans*. The percentage of infected flies was found to be 0.46. This is given as the minimum, and it is said that the real proportion is probably about three or four times as great.

It is also stated that given an outbreak in a herd there is some evidence to shew that *Tabanidae*, *Haematopota*, and other biting flies may act as mechanical carriers. The evidence that *Stomoxys* plays a similar rôle is said to be unsatisfactory.

- (17) BRUCE (David), HARVEY (D.), HAMERTON (A. E.), & Lady BRUCE. **The Trypanosome causing Disease in Man in Nyasaland.—Susceptibility of Animals to the Human Strain.**—*Proc. Roy. Soc.* 1913. Oct. 1. Series B. Vol. 87. No. B. 592. pp. 35–45.

The bulk of this paper is made up of tables shewing the results of experimental inoculation of different species of animals with trypanosomes derived from five human sources.

Ox. The trypanosome is not so deadly as *T. pecorum*. Of 13 animals inoculated only one died, after an illness lasting 134 days, four became infected, and the remainder proved refractory. The recovered animals are in good condition, presenting a contrast with animals recovered from *pecorum* infection.

Goat. Twenty-two animals were inoculated and all died. The duration of the disease varied from 19 to 72 days. One animal shewed swelling of the face, but none developed any opacity of the cornea.

Sheep. The infection runs about the same course in the sheep as in the goat, and the same fatal result is obtained. Of seven sheep inoculated three shewed marked swelling of the face.

Baboon. One was inoculated and proved refractory.

Monkeys. Twenty monkeys died in from 10 to 51 days. Trypanosomes were always present in their blood and were often very numerous. There was no oedema of the face, and no corneal opacity was observed.

Dog. Of 25 dogs inoculated all died in from 14 to 78 days. In eight of the animals there was oedema of the face and opacity of the cornea.

Rabbits. Seven died in from 15 to 42 days. The clinical picture of an infected rabbit is said to be exactly like that of one suffering from infection with nagana. There is swelling of the eyes, the face puffs up, and sores break out around the eyes and nose. The eyes become closed up and there is a purulent discharge from the nose. The ears become thickened and covered with sores.

Guinea-pig. These animals are more refractory than rabbits, and often require to be re-inoculated before they take the disease. Fifteen were used, and they died in from 12 to 114 days.

Rat. Twenty-one were inoculated and all died in from 13 to 93 days.

Among the conclusions drawn is the following:—

“It is not satisfactorily proved yet to what species this trypanosome belongs, but the Commission at present leans to the opinion that it is *T. brucei* (Plimmer and Bradford).”

- (18) DUKE (H. L.). **Some Trypanosomes recovered from Wild Game in Western Uganda.**—*Rept. of the Sleeping Sickness Commission of the Royal Society.* 1913. No. 14. pp. 37-59. With 4 plates and 1 map.

The tour of investigation dealt with in this paper was undertaken with the object of obtaining information regarding the part played by wild game in the spread of trypanosomiasis of human beings and domestic animals.

The area covered is divided roughly into four areas as follows :—

1. The north-west portion of Ankole.
2. Parts of the Toro Province from the northern bank of the Kazinga Channel to the Mahokya and Hima Rivers.
3. The Southern Toro Game Reserve, extending from the northern end of Lake George northwards nearly to Fort Portal.
4. The Semliki valley north of the Mboga road.

The characteristic topographical features and predominant game species of each of these districts are dealt with in some detail. A series of wild animals shot in districts where tsetse are not known to occur served as controls.

Blood from freshly killed animals was defibrinated and used for the inoculation of goats, this species being selected in order to include as many species of trypanosomes as possible. At the same time smears were taken for staining and fresh preparations were examined. The goats used were purchased in tsetse-free districts and were examined daily before use. No evidence of natural infection was found.

It was found impossible to set aside a goat for each species of game examined owing to the distances covered and the difficulties of transport.

Full details of every goat inoculated are given and it may be gathered that the following trypanosomes, or trypanosomes closely resembling them, were found :—*T. vivax*, *T. pecorum*, *T. uniforme*, a trypanosome very suggestive of *T. gambiense*, *T. nanum*, and a trypanosome belonging to the *brucei-pecaudi-gambiense* group.

Trypanosomes were found in the blood of one antelope only out of 39 animals of different kinds shot in districts known to be free of tsetse, but it is said that possibly this animal may at some time have reached the fly zone on Lake Victoria.

Of the 52 animals shot in the tsetse districts 10 were found to be infected with trypanosomes, while only one of those shot in the tsetse-free districts was found to be infected. Since tabanids, *Haematopota*, *Muscidae*, *Chrysops*, etc., are common to both districts, there can be no doubt regarding the significance of *Glossina* as a carrier of trypanosomes.

Details are given of a number of investigations carried out with wild *Glossina*.

Conclusions :—

" 1. A considerable portion of the wild game in the fly districts of the Western Province of the Uganda Protectorate is infected with trypanosomiasis.

" 2. Certain of these trypanosomes have suspicious resemblance to the human trypanosomes, *T. gambiense* and *T. rhodesiense*. Others are known to be exceedingly pathogenic to domestic animals.

" 3. The tsetse of these parts also contain flagellates which it is highly probable are derived from wild animals.

"4. The population of the fly districts is scanty, and the greater part has recently been moved to fly-free parts.

"5. It is reasonable to hope that, as regards the spread of human trypanosomiasis, the removal of the people from the infected districts will suffice, and this measure will simultaneously prevent native cattle being exposed to the fly bite. The majority of the fly area is practically uninhabited country.

"6. The alternative of destroying the game, and so of abolishing what is doubtless a permanent trypanosome reservoir, would be a gigantic and almost impossible undertaking in this region. In considering such a proposal elephant must, of course, be included, as must also hippo, situtunga, bush-pig and hyaena, all of which are difficult to eradicate. The scarcity of people is a serious objection to such a course, as is the difficult nature of the country. It would be well to await the trial of this expedient under more favourable conditions before undertaking so drastic a measure under severe natural handicaps.

"7. It would appear inadvisable to take any measures to protect the existing game and thereby encourage an increase in their numbers. On the other hand, it is inadvisable to permit natives to hunt in the fly districts. The ideal arrangement would be to make the fly districts prohibited areas, and in the region under consideration this is feasible to a far greater extent than would be the case in other parts of the Protectorate.

"Special attention must be directed to the Mahokya district in order to stamp out *G. pallidipes* from the immediate neighbourhood of the main road."

(19) BATTAGLIA (M.). Einige durch Trypanosomiasis Dromedarii erzeugte Läsionen. [Lesions in the Dromedary caused by Trypanosomes.]—*Centralbl. f. Bakt.* 1. Abt. Orig. 1913. Oct. 4. Vol. 71. No. 2/3. pp. 182-184.

The author claims to have established the fact that this trypanosome multiplies by schizogony and sporogony. The parasite was obtained from PRICOLA, who found it in a camel.

A rabbit was scarified with all possible aseptic precautions upon the prepuce with a hollow needle containing blood rich in trypanosomes. On the 4th day there was a hard, oedematous area at the seat of scarification, and by the 8th day there was scab-formation followed by ulceration and a hard granuloma. On the second day after scarification there were present in the blood intra- and extraglobular amoeboid forms of the parasite. On the 8th day, when true trypanosome forms were scantily present in the blood, rounded and pear-shaped forms were numerous present. Trypanosome forms were observed in large numbers in the lesions of the genital organs. There was oedema of the scrotum and orchitis.

In rabbits infection with this trypanosome runs a peculiar course. Infection occurs in whatever manner the animals are inoculated. During the first three days trypanosome forms are scantily present in the blood and after this they disappear although the blood remains infective. The animals gradually lose condition and die in from seven to eight months.

The local lesion of the genital organs is not developed if the inoculation is made at some other part of the body.

Rabbits and dogs sometimes shew keratitis.

- (20) FAVERO (Francesco). **Su alcune Vie d'Introduzione del *Trypanosoma brucei*, considerate in Rapporto al Decorso dell'Infezione.** [On the Different Channels of Introduction of *Trypanosoma brucei*, considered in Relation to the Course of the Infection.]—*Clinica Veterinaria*. 1913. Nov. 30. Vol. 36. No. 22. pp. 996-1002, & Dec. 15. No. 23. pp. 1045-1054. With 1 chart.

The author, having set himself to discover what difference resulted from inoculating *Trypanosoma brucei* into different regions in dogs, found that the intensity of the resulting infection varied according to the seat of the injection, in the following order:—1, intra-thyroid, 2, intra-renal, 3, intravenous, 4, intra-urethral, 5, intra-testicular, 6, endo-peritoneal, 7, subcutaneous, 8, intra-medullary, 9, intra-splenic, and 10, into the anterior chamber of the eye. The results are exhibited in tabular form. The aqueous humour of the eye seems to have a distinctly attenuating effect upon *Trypanosoma brucei*, to judge from the mildness of the resulting infection.

The greater part of the paper is occupied with a discussion of the views of various authorities, and there is a copious bibliography at the end.

- (21) GORETTI (G.). **Ricerche Sperimentali sul Nagana. III Comunicazione. Contributo allo Studio delle Alterazioni del Sistema nervoso centrale nell'Infezione sperimentale da Nagana (*Trypanosoma brucei*).** [Experimental Investigations regarding Nagana. III Communication. The Alterations in the Central Nervous System in Experimental Nagana (*T. brucei*).]—*Lo Sperimentale*. 1913. Nov. 13. Vol. 67. No. 5. pp. 527-564. With 2 plates.

The author's conclusions are as follows:—

In rabbits, guinea-pigs and dogs experimentally infected with *Trypanosoma brucei* (nagana) serious lesions are found in the central nervous system.

1. Marked chromatolysis of the cellular elements in general.
2. Moderate infiltration with plasma cells and lymphocytes in the membranes and around the capillaries of the central nervous system, similar cells being found free in the lumen of the vessels. Also a moderate scattering of "Stäbchenzellen" in the nervous tissues, particularly in the case of dogs.
3. Diffuse lesions of the myeline fibres of the brain, some of the tracts of the cord, the bulb, pons, mesencephalon and of the roots of the spinal and cranial nerves. These alterations can be demonstrated by the methods of DONAGGIO and MARCHI.
4. More advanced bilateral lesions of the decussating pyramidal tracts may be demonstrable by WEIGERT's method, and the posterior roots and posterior spinal tracts may also be involved.
5. A slight proliferation of the neuroglia.
6. The lesions are in part of a secondary nature and probably due to toxic products liberated by the trypanosomes. Such products have already been shown to have an injurious effect upon the central nervous system.

- (22) PAPARONE (E.). **Ricerche Sperimentali sul Nagana. IV Comunicazione. Lesioni oculari per Infezione generale da *Trypanosoma brucei*.** [Experimental Investigations regarding Nagana. IV Communication. Ocular Lesions in General Infection with *T. brucei*.]—*Lo Sperimentale*. 1913. Dec. 17. Vol. 67. No. 6. pp. 933-942. With 1 plate.

The experiments given in this paper were carried out with a very

virulent strain of the trypanosome, mice dying in three days after intraperitoneal inoculation. It was found that in mice and rats death occurred in so short a time that no ocular lesions visible to the naked eye were produced. Lesions were found in rabbits and dogs, which offered more resistance to the infection and consequently survived for a longer period after inoculation. In from a week to a fortnight these animals showed swelling of the eyelids and adjacent tissues and a scabby condition of the edges of the lids. There was also a more or less intense conjunctivitis with a variable amount of muco-purulent secretion. The infected rabbits and dogs, and particularly the latter, showed a parenchymatous keratitis of the cornea associated with iritis.

The conclusions drawn are :—

1. In dogs and rabbits experimentally infected with *T. brucei* lesions may be observed involving the tissues of the eye. Blepharitis, conjunctivitis, intense diffuse parenchymatous keratitis, irido-cyclitis, choroiditis, etc., may be seen.
2. The trypanosomes may be found in the conjunctival secretion and in the aqueous humour (in stained preparations and by animal inoculation).
3. They are demonstrable in the cornea in the earliest stages when it is still transparent and when the keratitis is intense. The interstitial lesions are due to the multiplication of the parasites in the parenchyma of the cornea and to their toxic products. It has been found possible to produce parenchymatous keratitis with extracts of trypanosomes.
4. As the aqueous humour contains trypanosomes, whatever the method of infection, so animals can be infected by inoculation into the anterior chamber of the eye.

- (23) BOVONE (E.) & MAJA (A.). Nota su di un Tripanosoma dei Bovini del Katanga. [A Trypanosome of Bovines in Katanga.]—*Giorn. di Med. Vet.* 1913. Oct. 25. Vol. 62. No. 43. pp. 1033–1035.

The authors describe the occurrence of a large trypanosome in the blood of seven bovines on the Esschen Plateau. In fresh blood the parasite was exceedingly active, the movement involving translation. The nucleus was visible and appeared to be about the size of a red blood corpuscle, and the posterior portion of the body shewed a number of large granules. Although the centrosome could not be made out, it appeared to be placed near to the nucleus, and the undulating membrane and flagellum took origin from that situation. The parasite was very scantily present in the blood.

Only one stained preparation was available for examination and in this the parasite measured 60 microns in length by 5 in width. Both ends tapered to a marked degree. The authors think that the organism was *T. ingens*.

- (24) FERBER (F.). Beiträge zur Biologie der nur auf kulturellem Wege nachweisbaren Flagellaten des Rinderblutes. [The Biology of the Culturally Demonstrable Flagellates of Ox Blood.]—*Zeitschr. f. Hyg. u. Infektionskrankh.* 1913. Dec. 12. Vol. 76. No. 2. pp. 193–207,

Multiplication of the parasites is inhibited either partially or completely at 37° C., and at 53° C. the parasites gradually die. Temperatures of 50° C. and lower for a period of 24 hours do not injure the vitality of the parasite.

The flagellate multiplies in broth prepared from various kinds of meat, but not in defibrinated blood or salt solution.

The parasite multiplies on faintly alkaline agar and on blood agar, and in blood broth whether the blood used for the medium is taken from an animal that has been infected or not.

In no stage of development can the flagellates pass Berkefeld filters.

It was found to be impossible to infect animals with cultures, as indicated by the non-appearance of trypanosomes in the blood and the absence of flagellates in cultures.

- (25) BLACKLOCK (B.) & YORKE (W.). *Trypanosoma vivax* in Rabbits. —*Ann. Trop. Med. & Parasit.* 1913. Dec. Vol. 7. No. 4. pp. 563-568.

The strain of *T. vivax* used in this series of experiments was that isolated by the authors from a horse from the Gambia (see this *Bulletin*, Vol. 1. No. 2. Abstr. No. 121. pp. 76-77). After the isolation of the parasite the strain was preserved by passage through goats, the number of passages up to the date of writing being 39. During this period there was observed no exaltation of virulence for the goat. The average duration of the disease during the whole period has been 30 days.

A number of attempts were made to infect rabbits from the goats of this series, but the inoculations failed or resulted in a transitory infection from which the animals recovered. From the 38th goat of the series, however, rabbits were successfully infected by intraperitoneal inoculation.

The period of incubation was 5 to 8 days, and of the four rabbits used two died, in 17 and 34 days and the other two recovered.

From these rabbits the strain has been carried on in others over a period of more than three months to the eighth generation. Goats have been successfully inoculated from rabbits. The strain has not proved to be invariably pathogenic for the rabbit, and many of them appear to have recovered, but the majority have certainly become infected.

¶ Examination of the strain after passage through six generations in the rabbit shewed that the parasite as a rule did not exhibit the same degree of activity as in the goat, but there were no discoverable alterations of morphology.

- (26) MESNIL (F.). Sur le Nagana de l'Ouganda.—*Bull. Soc. Path. Exot.* 1913. Dec. Vol. 6. No. 10. pp. 685-689.

After referring briefly to the literature regarding the identity or otherwise of *T. brucei* Plimmer and Bradford and *T. brucei* (Uganda), the author gives details of some experiments carried out with the idea of throwing further light upon the question.

Three goats, two of which had been used for experimentation with other trypanosomes, were inoculated with the Uganda strain, the "ferox" (Ehrlich) nagana strain and the Zululand strain. The first and last of these died as a result of the inoculations. The goat inoculated with the "ferox" strain survived and apparently recovered, since its blood was non-infective for a dog. It received a second inoculation with the same strain and became reinfected. Before it had recovered from the second inoculation it was inoculated with

the Zululand strain of nagana. About six months after this inoculation its blood was negative when tested by the inoculation of dogs. After a further interval of six months it was again inoculated with the Zululand strain and failed to become infected, as shewn by the experimental inoculation of mice.

The author concludes that at this date the goat was immune to the Zululand strain.

Two months later it was inoculated with the Uganda strain, and succumbed to the infection after an interval of two months.

From this the author concludes that the Zululand strain is different from the Uganda strain, and this view is supported by the results obtained in some passive immunity experiments which are briefly referred to.

- (27) MACFIE (J. W. Scott) & JOHNSTON (J. E. L.). A Case of Equine Trypanosomiasis characterised by the Occurrence of Posterior Nuclear Forms.—*Jl. Trop. Med. & Hyg.* 1913. Nov. 15. Vol. 16. No. 22. pp. 348-349. With 1 text figure.

The trypanosomes described in this short note were found in films of blood taken from a horse at Accra. Long slender and short stumpy forms were met with and in many cases the tropho-nucleus was situated in the posterior half of the body. In no instance was a trypanosome observed in which the tropho-nucleus was actually posterior to the kineto-nucleus, although in many it was touching it.

Following the procedure described by BLACKLOCK the authors found that the nucleus was definitely posterior in 4·8 per cent. of all trypanosomes, except dividing forms, present.

As it was impossible to carry out any inoculation experiments the authors are unable to identify the parasite with certainty, but they state that a parasite with the same morphology is responsible for the death of a large number of horses in Northern Nigeria every year. Several specimens were identified by BRUCE as *T. brucei*.

The author points out that, if *T. brucei* (*pecaudi*) is identical with *T. rhodesiense*, one would expect to find cases of human trypanosomiasis due to *T. rhodesiense* in places where *T. brucei* is common among the domesticated animals. This is not the case in Nigeria. Sleeping sickness there does not present the acute features observed in cases in Rhodesia. The disease is endemic, but by no means fatal.

One of the authors has recently studied the trypanosome from a case of sleeping sickness in Southern Nigeria, and it is stated that neither in its morphology, measurements, nor in its animal reactions does it resemble *T. brucei* (*pecaudi*) or *T. rhodesiense* (see below p. 25).

- (28) NEIVA (A.). Multiplicacao no Vinchuca (*Triatoma infestans* Klug) do Trypanosomo do Mal de Cadeiras. [The Multiplication of the Trypanosome of Mal de Caderas in the Vinchuca.]—*Brazil Medico.* 1913. Sept. 15. Vol. 27. No. 35. pp. 366-367.

The author has been unable to find a correspondence in distribution between the capybara and mal de caderas, although it was suggested by ELMASSIAN and MIGONE that that animal was the reservoir for the trypanosome. He has been able to infect the vinchuca experimentally

and has found that its faeces are infective for guinea-pigs when placed upon the conjunctiva. The period of incubation was 10 to 15 days.

It is suggested that flies of the genus *Chrysops* are more probably the transmitting agents than *Stomoxys calcitrans*, as suggested by SIVORI and LÉCLER.

- (29) BLACKLOCK (B.) & YORKE (W.). The Trypanosomes causing Dourine (Mal de Coït or Beschälseuche).—*Proc. Roy. Soc.* 1913. Oct. 16. Series B. Vol. 87. No. B. 593. pp. 89-96. With 1 plate.

In this paper the authors record the results of their examinations of three strains of trypanosomes said to be the trypanosome of Dourine. The first of these was obtained from the Kaiserliches Gesundheitsamt, Berlin, and is said to have come originally from Algeria, the second from EHRLICH, the origin of which is unknown, and the third was the strain responsible for the outbreak of dourine in Eastern Prussia in 1905.

The biometrical method was adopted.

The authors find that the second and third strains closely resemble each other morphologically, while the first presents marked differences. The second and third belong to the group of trypanosomes in which all individuals have a free flagellum, while the morphology of the first strain does not enable them to distinguish it from *T. rhodesiense* or *T. brucei* Uganda.

They are conducting experiments to ascertain whether the strains are still capable of setting up the typical disease in equines. For the present they propose the name *T. equi* for the *rhodesiense*-like organism.

- (30) BAUCHE (J.) & BERNARD (P. N.). Notes sur le Surra d'Indo-Chine à Hué.—*Bull. Soc. Path. Exot.* 1913. Dec. Vol. 6. No. 10. pp. 690-693.

The natural disease.—Equine trypanosomiasis in Annam has been differentiated from the surra of India by LAVERAN and MESNIL, by whom the causal trypanosome was named *T. annamense*. The disease exists in Annam as an enzootic among horses, cattle, buffaloes, and dogs. In a paper published by SCHEIN in 1907 the part played by cattle in forming the reservoir of the virus was emphasised.

The authors have examined the blood of 300 cattle at the slaughter-houses in apparent good health and have discovered trypanosomes by direct observation on six occasions. Eighty-nine buffaloes were examined with negative results. A single infected dog was found among 300 examined.

In two outbreaks which resulted in the death of 9 out of 48 and 100 out of 200, the horses were stabled near the cattle sheds. The batches of horses pastured near woods furnished the whole of those that became infected, while those that were pastured upon cleared land, going out only at night or on cool days when there was no sun, remained absolutely free from infection.

The average period of incubation was 11 days, and it was found possible to arrest an outbreak by taking daily temperatures, isolating

the animals shewing a febrile elevation and slaughtering them after a diagnosis by microscopic examination.

Experiments carried out with a variety of species of flies upon guinea-pigs failed.

Experimental inoculations.—It having been suggested by BALDREY that the pig might be the reservoir of surra, the authors examined the blood of about 2,000 pigs without discovering a single one infected. The trypanosome was found to be pathogenic for pigs, death taking place in from 10 to 75 days. Of a number of guinea-pigs inoculated from an infected dog the majority died in from three weeks to a month, and two monkeys died in 13 and 26 days.

In experimentally infected pigs the trypanosomes appeared in the peripheral blood in two to five days after intraperitoneal inoculation, and in from seven to eleven days after subcutaneous inoculation. The fever is intermittent and the appearance of trypanosomes in the blood coincides with the elevations of temperature. At the time of death the organisms are very numerous in the blood.

Clinically the disease is characterised simply by progressive emaciation, without any oedema, paralysis or lesions of the eyes.

At the post-mortem the spleen may be found to be three times the normal size.

- (31) MOHLER (J. R.), EICHORN (A.), & BUCK (J. M.). **The Diagnosis of Dourine by Complement-Fixation.**—*American Jl. Vet. Med.* 1913. Nov. Vol. 8. No. 11. pp. 581-587.

After trying a number of methods of obtaining antigen the authors adopted the plan of using an emulsion of the spleen of rats made with salt solution. The trypanosome used as antigen was not *T. equiperdum*, but *T. evansi*. "As dourine is the only known trypanosome infection of horses existing in this country, the value of even a group reaction was immediately appreciated and attention was directed to the carrying out of this idea in our diagnostic work."

Excellent results are claimed for the method.

- (32) BRUCE (David), HARVEY (D.), HAMERTON (A. E.), & Lady BRUCE. **Morphology of Various Strains of the Trypanosome causing Disease in Man in Nyasaland.—The Mzimba Strain.**—*Proc. Roy. Soc.* 1913. Oct. 1. Series B. Vol. 87. No. B. 592. pp. 26-35. With 3 plates.

This paper deals with the question whether the trypanosome causing human disease in Nyasaland is restricted to the game and "fly" of the Proclaimed area or whether it extends north and south along the fly-belt. The trypanosome dealt with was found in the blood of a donkey at Mzimba about 100 miles north of the northern border of the Proclaimed area. Tables of measurements of the trypanosome in a number of different animals, and three plates illustrating the fixed and stained organism are given. The conclusions arrived at are as follows:—

"1. The trypanosome of the Mzimba strain is the same species as that occurring in the wild game inhabiting the Proclaimed area, Nyasaland.

"2. It has already been concluded that this species is *T. brucei* vel *rhodesiense*.

"3. Hence it would appear that wild *G. morsitans* occurring in a district 100 miles north of the Proclaimed area are infected with the trypanosome which causes the human trypanosome disease of Nyasaland."

(33) ECKARD (B.). Uebertragung des *Trypanosoma rhodesiense* durch die *Glossina palpalis*. [The Transmission of *T. rhodesiense* by *G. palpalis*.]—*Centralbl. f. Bakt.* 1. Abt., Orig. 1913. Nov. 26. Vol. 72. No. 1/2. pp. 73-76.

In this paper the author gives a chronological list of the more important work that has been done in connection with the transmission of trypanosomes by tsetse flies and draws the conclusion that given favourable conditions any known pathogenic trypanosome can develop in any species of *Glossina*. Further observations are necessary to decide whether any particular species of *Glossina* is especially concerned in the transmission of trypanosomiasis.

The author has had the opportunity of carrying out some experiments in the transmission of *T. rhodesiense* derived from a case of sleeping sickness in Nyasaland with *G. palpalis*.

Four hundred and seventy-six laboratory-bred *G. palpalis* were fed in series on infected guinea-pigs and after a period of starvation of two days were fed upon healthy goats and monkeys. Some of these animals were found to be infected on the 32nd day.

It was found that 2.5 per cent. of the total number of flies were infective, and developmental forms were found in nine flies which had died by the 10th day.

Three of the infective flies were used for the purpose of determining the infectivity of the trypanosomes in the various organs. After they had been fed upon healthy animals for two days they were killed by chloroform and their salivary glands, proventriculus and intestines were used for the subcutaneous inoculation of healthy monkeys. All the animals were found to be infected on the 5th day.

These results are in agreement with those obtained by KINGHORN and YORKE, but in striking contrast with those obtained by KLEINE, ECKARD, and Miss ROBERTSON.

The possibility is considered that injury to the salivary glands might have been responsible for the contamination of the other organs, but the author is of the opinion that the number of experiments negatives this possibility.

It is further suggested that the ages of the flies might be a determining factor.

In the author's experiments the flies were 40 days old and in those of KLEINE and ECKARD they were from 50 to 60 days old. With the object of obtaining some evidence on this point a fly was kept until the 54th day when it was dissected, its organs being used for the inoculation of monkeys. In this case only the one inoculated from the salivary glands became infected. Examination of the fly shewed that numerous trypanosomes were present in the intestine, and the author concludes from this that development in the intestine gradually declines, the virulent parasites making their way to the salivary glands.

- (34) KLEINE (F. K.) & ECKARD (B.). Ueber die Bedeutung der Haustiere und des Wildes für die Verbreitung der Schlafkrankheit. [The Importance of Domesticated and Wild Animals in the spread of Sleeping Sickness.]—*Zeitschr. f. Hyg. u. Infektionskrankh.* 1913. July 17. Vol. 75. No. 1. pp. 118-122.

Experiments were carried out in which infective *G. palpalis* were fed upon healthy sheep, goats and monkeys. Of 13 instances in which sheep and goats were used the result was negative in nine, while in the monkeys (number not stated) the result was invariably positive.

The authors were unable to infect *G. palpalis* on three out of seven animals in whose blood *T. gambiense* was to be found.

Two goats which had been infected in September 1910 recovered, as shewn by experimental inoculation and fly experiments. The animals failed to become infected again when re-inoculated subsequently.

Five oxen, 55 goats and 25 sheep from districts in which sleeping sickness was widely spread were examined and *T. gambiense* was found in one ox, one goat and one sheep.

The authors failed to find *T. gambiense* in any of the game examined.

- (35) TAUTE (M.). Untersuchungen über die Bedeutung des Grosswildes und der Haustiere für die Verbreitung der Schlafkrankheit. [The Significance of Big Game and Domesticated Animals in the Spread of Sleeping Sickness.]—*Arbeit. a. d. Kaiserl. Gesundheitsamt.* 1913. Aug. Vol. 45. No. 1. pp. 102-112. With 1 plate.

These experiments were carried out at Lubimbinu in Portuguese Nyasaland. This place is itself fly-free, but is surrounded by a fly belt in which *G. morsitans* is plentiful. The population is free from trypanosomiasis, but the contrary is the case with domestic animals. Cattle and sheep are not found, goats and dogs only occasionally.

Thirty-seven wild animals were examined and trypanosomes were found in 20, either by direct examination or by experimental inoculations. Trypanosomes possessing the pathogenic range of *T. rhodesiense* were found in six of the inoculated monkeys and dogs. The remaining trypanosomes found belonged either to the *T. vivax* or *T. pecorum* groups.

Polymorphic trypanosomes shewing posterior nuclear forms were found in six out of ten animals upon which wild *G. morsitans* were fed. Seven dogs and a goat from the fly-belt were found to be infected with a trypanosome which was morphologically similar.

Experimentally infected dogs and goats died on an average in 36 and 16 days respectively, and shewed keratitis and oedema of the face.

The author states that if the trypanosome be in reality *T. rhodesiense*, about 16 per cent. of the game and domesticated animals are infected while the population remains free from infection.

To test this question of identity the author fed a number of laboratory-bred flies upon a monkey infected with the antelope strain. After a period of starvation for a day they were fed upon a series of goats, monkeys and dogs up to the 27th day. From the 28th to the 32nd day they were fed upon the author and from the 33rd to the 44th day upon another series of animals. The four animals upon which

the flies were fed from the 20th to the 27th day became infected, as did the three animals fed upon from the 33rd to the 44th day, but the author remained in good health.

A second experiment of a similar nature was carried out and with the same result.

In a third experiment the author was inoculated with 2 cc. of blood from a naturally infected dog but failed to become infected, while animals inoculated with the same blood all became infected and died.

Blood drawn from the author failed to infect a monkey and a number of dogs.

The author considers that the trypanosome of game and domesticated animals is *T. brucei*.

- (36) MELLO (U.). *Le Trypanosoma gambiense a-t-il une Affinité pour le Testicule?*—*Bull. Soc. Path. Exot.* 1913. Oct. Vol. 6. No. 8. pp. 583-588.

The experiments detailed in this paper were carried out to control the statement made by UHLENHUTH and EMMERICH to the effect that in rabbits inoculated with *T. equiperdum* and *T. gambiense* the multiplication of the trypanosomes is far more advanced in the testicles than in the blood. This multiplication takes place, according to these authors, in both testicles even when only one has been the seat of inoculation, and it occurs even when the animal has been inoculated by some other path.

Seven rats, five guinea-pigs, four rabbits, and a monkey were used, and the trypanosome employed was *T. gambiense*.

Rats. In rats the trypanosome first made its appearance in the blood, no matter in what manner the animal was inoculated. Parasites could be found in small numbers only in the testicle, even when that had been the seat of inoculation, and they were always more numerous present in the blood.

Guinea-pigs. Trypanosomes made a rapid appearance in the testicles in those cases only in which that had been the seat of inoculation. After intra-testicular inoculation the parasites may be discoverable in the other testicle before they can be found in the blood.

Monkey. The appearance of trypanosomes in the testicle did not precede their appearance in the blood.

Rabbits. The experiments carried out with these animals at first offer the most support to the view expressed by UHLENHUTH and EMMERICH, since on several occasions trypanosomes were fairly numerous in the testicles while still scanty in the blood (as is the rule in rabbit trypanosomiasis). The author does not think that this is conclusive. He was informed by LAFERAN that he had frequently observed that trypanosomes multiply more rapidly in oedematous liquid when that is present in any part of the body than in the blood, and it was in the oedema resulting from the inoculation or from repeated punctures that the trypanosomes were found. Further, when a pipette is introduced into the testicle the liquid obtained is frequently from the sheath of the testicle and not from the substance of the organ itself. It is easy to conceive that trypanosomes might be more numerous in this than in the blood after intraperitoneal inoculation.

- (37) MACFIE (J. W. Scott). On the Morphology of the Trypanosome (*T. nigeriense*, n. sp.) from a Case of Sleeping Sickness from Eket, Southern Nigeria.—*Ann. Trop. Med. & Parasit.* 1913. Aug. Vol. 7. No. 3A. pp. 339–356. With 1 plate.

In the unstained state it can be observed that some of the parasites are long and slender and others short and stout. The former execute rapid vibratory and translatory movements. The translatory movement is spasmodic.

In preparations fixed with alcohol or osmic acid and stained with Giemsa the longest trypanosome found measured 32 microns and the shortest 8 microns. The breadth varies from 1 to 2.5 microns, and the short individuals are generally the stoutest. The trypanosome is polymorphic. Two long slender forms have been observed, one in which the micronucleus is terminal and the other in which it is situated about 1 micron from the posterior extremity, the extremity of the body being drawn out into a blunt “snout.” In some parasites the nucleus shews a clear area in the centre.

The trypanosome appears to be feebly pathogenic for laboratory animals. The original guinea-pig in which the author received the strain had at the time of writing been alive 124 days since inoculation and was still apparently well. Other animal inoculations made so far are as follows:—Five guinea-pigs have not shewn trypanosomes and one of these has been inoculated 56 days. Three mice out of seven have shewn very small numbers of trypanosomes in from four to eight days after inoculation. Two goats and two dogs have never shewn trypanosomes, and in one monkey they appeared in small numbers on the 10th day, but have up to the time of writing (22 days after inoculation) always been scanty.

- (38) MACFIE (J. W. Scott). Preliminary Note on the Development of a Human Trypanosome in the Gut of *Stomoxys nigra*.—*Ann. Trop. Med. & Parasit.* 1913. Nov. Vol. 7. No. 3B. pp. 359–361. With 1 text-fig.

In this short note the author records the results obtained in a small number of experiments which circumstances compelled him to interrupt.

A number of *Stomoxys nigra* were fed upon a guinea-pig infected with a trypanosome from a case of sleeping sickness from Eket in Southern Nigeria. Thirteen flies were dissected from one to six days after being fed. In six of them flagellates were found in the mid-gut. Twelve flies caught in the laboratory which had not been fed upon the guinea-pig were dissected, but in none were any flagellates found.

Further experiments were made with flies bred in the laboratory. Two bred flies were fed upon the infected guinea-pig the day after hatching, and three days later herpetomonas were found in the mid-gut of each. There were none in the salivary glands or the proboscis. As controls two bred flies which had not been fed upon the guinea-pig were dissected and no flagellates were found.

- (39) LAVERAN (A.). **Au Sujet des Infections des Souris par le *Trypanosoma duttoni*.** [The Infection of Mice with *T. duttoni*.]—*Bull. Soc. Path. Exot.* 1913. Nov. Vol. 6. No. 9. pp. 626-627.

The author received six mice inoculated with the trypanosome from Senegal. The species of mouse was *Mus maurus*. They were all shewing trypanosomes in their blood at the time of arrival. Four of these mice were killed for the inoculation of white mice, and the infection was followed by periodical examinations in the other two.

The author was unaware how long the mice had been infected before they came into his possession, but after their arrival they lived for 22 and 26 months and apparently died of old age. Trypanosomes could always be found in their blood in larger or smaller numbers.

Two of the white mice inoculated appeared to make complete recoveries, trypanosomes disappearing from their blood after seven and four and a half months respectively.

It was shewn by inoculation that the progeny of infected mice possessed no immunity.

- (40) LAVERAN (A.). **Trypanotoxines. Essais d'Immunisation contre les Trypanosomes.**—*Bull. Soc. Path. Exot.* 1913. Dec. Vol. 6. No. 10. pp. 693-698.

The author has repeated with slight modifications the experiments of SCHILLING and RONDONI regarding the production of trypanotoxines in liquids containing dead trypanosomes, when these are heated for certain periods at 37° C., and the immunising effects of sub-toxic doses of such materials. He has failed to produce either the toxic or the immunising effect, and concludes with regard to the latter that when the results are not entirely negative they are partial and inconstant.

- (41) LEVADITI (C.) & MUTERMILCH (St.). **Recherches sur la Production des Anticorps chez les Animaux Trypanosomiés et traités par le Salvarsan.**—*Bull. Soc. Path. Exot.* 1913. Dec. Vol. 6. No. 10. pp. 699-704.

The authors' experiments in connection with this subject, which have not been previously published, were carried out in 1911, and the reason for their publication lies in the publication of the results obtained by MARGULIES, and ELLIS and SWIFT.

They find that if "606" be administered intraperitoneally to rats infected with nagana, on the third day of infection, when the trypanosomes are numerous in the blood, the serum becomes trypanocidal two hours later. The longer the period elapsing between the disappearance of the trypanosomes from the blood and the taking of the sample of serum the less is its trypanocidal power. This power disappears completely by the 30th hour. The fact that heating the serum to 56° C. does not destroy its trypanocidal power indicates that it is not due to trypanolytic properties of the serum, but to the presence of "606" in it.

On the other hand evidence is furnished by the experiments that, in spite of the richness of the blood in trypanosomes and the considerable quantity of antigen liberated by the action of the drug, antibodies do not make their appearance until the 5th day after inoculation.

- (42) SCHILLING (C.) & RONDONI (P.). **Tossine Tripanosomiche e Immunità di fronte ai Tripanosomi.** [Trypano-toxins and Immunity against Trypanosomes.]—*Lo Sperimentale*. 1913. Nov. 13. Vol. 67. No. 5. pp. 595-613.

In this paper the authors record the toxic effects of inoculating mice with suspensions of trypanosomes kept at 37° C. for variable periods.

They also found that emulsions of trypanosomes heated to 37° C. for certain periods were capable of conferring a degree of immunity.

- (43) JOHNSTON (J. E. L.) & MACFIE (J. W. Scott). **Observations on the Action on Trypanosomes of certain Drugs and of *Staphylococcus pyogenes*.**—*Jl. London School Trop. Med.* 1913. Nov. Vol. 2. No. 3. pp. 207-212.

This paper contains the records of a few investigations carried out with bromine, iodine, and osmic acid and also *Staphylococcus pyogenes*. The experiments were made *in vitro* and *in vivo*, but the results were almost entirely negative in character.

A few initial experiments were made to determine the effects of dilute solutions of iodine, bromine, and osmic acid upon trypanosomes in cover-slip preparations. The results obtained were very inconclusive.

In the experiments carried out on living animals, sheep infected with *T. vivax* and rats infected with *T. lewisi* were used. In the case of the sheep the injections were given intravenously, and in the rats subcutaneously. The solutions used were:—Tincture of iodine in distilled water in the proportion of 7 minims to the drachm, 2·5 per cent. bromine in the same proportion, and 2 per cent. osmic acid, 12 minims to the drachm. The rats received doses varying from 5 to 20 minims, and the sheep, of which only one appears to have been subjected to experiment, was given one drachm of pure tincture of iodine intravenously. The drugs appeared to have a disturbing effect not only on the trypanosomes but also on the hosts. The authors consider that trypanophagocytosis occurs and that large numbers of trypanosomes may be rapidly destroyed by this means.

The injection of staphylococcus vaccines into rats infected with *T. lewisi* appeared to be without effect.

- (44) MACFIE (J. W. Scott) & JOHNSTON (J. E. L.). **Auto-Erythrophagocytosis as an Aid to the Diagnosis of Trypanosomiasis. (Preliminary Communication.)**—*Jl. London School Trop. Med.* 1913. Nov. Vol. 2. No. 3. pp. 212-215.

The authors have observed the ingestion of red cells by leucocytes to which attention was drawn by CONNALL and others. They have made observations using *T. lewisi*, a trypanosome resembling *T. gambiense* from a case of sleeping sickness, and *T. duttoni*. They divide their cases into the following three groups:—

1. Those in which leucocytes containing red cells were present in blood which subsequently contained trypanosomes.

2. Those in which leucocytes containing red cells and trypanosomes were present at the same time.

3. Those in which leucocytes containing red cells were found after the disappearance of trypanosomes.

The ratio of cells containing red corpuscles to other leucocytes is seldom higher than 1 per cent.

The suggestion is put forward that erythrophagocytosis may be a material aid in the diagnosis of trypanosomiasis.

- (45) KOLLE (W.), HARTOCH (O.), ROTHERMUNDT (M.), & SCHÜRMANN (W.). **Chemotherapeutische Experimentalstudien bei Trypanosomeninfektionen.** [Experimental Chemotherapeutic Investigations in connection with Trypanosome Infections.]—*Zeitschr. f. Immunitäts. u. experim. Therapie.* 1913. Aug. 5. Vol. 19. No. 1. pp. 66-97.

This paper contains a repetition of results previously published (see References 194 and 556, Vol. 1, this *Bulletin*) together with a description of some further experiments with antimony trioxide.

Attention is drawn to the necessity of a thorough study of the actions of various compounds of antimony, as in the past it has been compared too closely with arsenic.

Experiments were carried out using metallic antimony reduced to a fine powder and suspended in oil. This material was injected intramuscularly into mice infected with nagana.

The strain of trypanosomes used was invariably fatal in five days, and the drug was administered on the 2nd or 3rd day.

It was found that the toxic dose was 5 mg. per 10 g. body weight, and that 1 mg. caused a disappearance of trypanosomes with the utmost certainty, and no relapses occurred. Larger doses invariably produced poisoning.

Numerous organic and inorganic compounds of antimony were tested with the object of finding out the connection between their composition and their therapeutic effects. Pentavalent compounds were found to be by far less toxic than the trivalent but they were practically without therapeutic effect in trypanosome infections. The trivalent compounds were both toxic and trypanocidal, particularly the trioxide, to which the authors give the name "Trixidin."

Experiments showed that a dose of 120 mg. per 10 g. body weight produced neither acute nor chronic toxic effects, nor did it produce any changes in the blood or organs.

It was observed that the intramuscular injections of large doses of "Trixidin" into rabbits, dogs, and apes was liable to lead to abscess formation. Experiments are being carried out with the object of avoiding this by distributing the injections over different parts of the body.

"Trixidin" is insoluble in water and is slowly absorbed.

While mice withstand the injection of 100 mg. per 10 g. body weight, one hundredth part of this quantity is effective in the treatment of experimental nagana in them.

The injection of sufficiently large doses of "Trixidin" protected mice in the majority of cases when inoculated as much as 16 days after with nagana and up to the time of writing this immunity had lasted 91 days.

A list is included in the paper shewing a number of compounds of various metals which were tried and which were found to be either useless, or, though useful, impracticable in application.

The authors have been able to effect cures with drugs, which when injected either lead to intoxication or permit of relapses, by applying them externally in the form of salves.

"Trixidin" was found to be ineffectual in the treatment of mice infected with the spirochaete of African relapsing fever and *Schizotrypanum cruzi*.

- (46) MONFORT (F.). *Essais de Traitement des Trypanosomiasés expérimentales par l'Arsénophénylglycine.*—*Bull. Soc. Path. Exot.* 1913. Oct. Vol. 6. No. 8. pp. 588-590.

The author's experiments have been carried out with adult mice inoculated with *T. gambiense*, *T. rhodesiense*, *T. dimorphon*, and *T. congolense*.

In the majority of cases the dose employed was 3 mg., but mice tolerate twice or three times this dose without shewing any symptoms. The drug was administered by intramuscular injection. Brief details are given of four series of experiments made with the trypanosomes mentioned and the results arrived at may be summarised as follows:—

In all, fourteen animals inoculated with the two human trypanosomes were treated and in no case was there a relapse, the animals being kept under observation up to 86 days in the case of the *gambiense*, and 64 days in the case of the *rhodesiense*.

Of five animals infected with *T. dimorphon* only one shewed a relapse during a period of 75 days.

Seven mice inoculated with *T. congolense* were put under treatment with doses of 3, 6, and 9 mg., but in no instance did the drug have any effect beyond a brief disappearance of the trypanosomes from the blood.

- (47) HECKENROTH (F.) & BLANCHARD (M.). *Le Néosalvarsan dans le Traitement de la Trypanosomiase humaine.*—*Bull. Soc. Path. Exot.* 1913. Oct. Vol. 6. No. 8. pp. 591-512.

In this paper the authors publish the results obtained in the treatment of human trypanosomiasis with a single intravenous injection of neosalvarsan. In 16 cases the persons treated were in good condition and in the remainder in moderate or bad condition.

The drug was administered dissolved in distilled water in the proportions of 0.15 g. and 0.45 g. per 25 cc. The dose varied from 1 cg. per kilog. to 2.1 cg. per kilog. It was observed in some cases in which the dose was 1.5 cg. or over that fever, vomiting, and diarrhoea occurred some hours after.

The drug appears to cause a rapid disappearance of the trypanosomes from the blood, but acts with less certainty upon those in the lymphatic glands. Trypanosomes have been discovered by gland puncture from 1 to 7 days after the administration of 1.7 cg. per kilog. of the drug.

Of the 25 cases treated 4 have not yet been re-examined, 9 have remained without relapse after an average period of $4\frac{1}{2}$ months, 2 died after 3 months and $1\frac{1}{2}$ months without relapse, and in 10 cases there has been a relapse after an average period of $2\frac{1}{2}$ months.

- (48) NÄGLER (K.). Experimentelle Studien über die Passage von *Schizotrypanum cruzi* Chagas durch einheimische Tiere. Teil I. [The Passage of *Schizotrypanum cruzi* through Indigenous Animals.]—*Centralb. f. Bakt.* 1. Abt., Orig. 1913. Oct. 4. Vol 71. No. 2/3. pp. 202-206. With 1 plate.

The author's conclusions are as follows:—

1. Mice, rats, guinea-pigs, rabbits, dogs, and cats are susceptible to infection with *Schizotrypanum cruzi*. It has not been found possible to transmit the parasite to canaries, lizards, and frogs.
2. The parasite is not transmitted hereditarily.
3. Infection experiments with insects failed.
4. The "male" and "female" forms described by CHAGAS as sexual individuals are vegetative forms.
5. No morphological differences are to be observed in the parasite obtained from mice, rats, and guinea-pigs.
6. In the peripheral blood, and even two hours after death, numerous divisions occur.

SPIROCHAETOSIS.

- (49) MARCHOUX (E.) & COUVY (L.). Argas et Spirochètes. Deuxième Partie. Le Virus chez l'Acarien. [Argas and Spirochaetes. Second Part. The Virus in the Tick.]—*Ann. Inst. Pasteur.* 1913. August. Vol. 27. No. 8. pp. 620-643.

The authors recapitulate the facts which they claim to have established in the first portion of this paper (see this *Bulletin*, Vol. 1. No. 5. pp. 298-301), viz., (1) the granules found in the tick stand in no relationship to the spirochaete and therefore the method of preservation of infection and hereditary transmission must be sought elsewhere; (2) spirochaetes retain their virulence in the coelomic cavity of the tick as such, with the single modification of a decrease in thickness.

In the second portion of the paper the authors propose to shew that spirochaetes alone are responsible for the transmission of the infection.

Experiments are quoted shewing that the inoculation of material obtained by grinding up infected ticks sometimes produces infection but more usually confers immunity. It is further shewn that inoculation with a portion of an infected tick such as a salivary gland or an oviduct is also followed by immunity.

That this immunity is not due to antibodies formed in the tick was shewn by the fact that if the material was heated to 55° C. for five minutes the power to infect was destroyed, while the power to confer immunity remained. On the other hand if the heating was continued for 20 minutes immunity did not result. These results agree with those obtained when material known to contain spirochaetes is treated in exactly the same way. It was also shewn that the filtration of such material destroyed its power both of infecting and immunising.

Using WRIGHT's method of estimating the number of spirochaetes present in a quantity of liquid, the authors have carried out a number of experiments having for their object the estimation of the number of spirochaetes required to confer immunity, and to produce infection. The bird selected for the experiments was the paddy and the following results were arrived at. By intramuscular inoculation it was necessary

to inject 20,000 spirochaetes in order to set up infection. Immunity was conferred when the number of spirochaetes injected lay between 17,000 and 735. If a smaller dose was administered no effect of any kind was produced. When intravenous inoculation was resorted to it was found that 17,000 spirochaetes caused infection, 70 conferred immunity, and 17 produced no effect. Thirty spirochaetes introduced into a fowl's egg during incubation sufficed to set up infection. The figures given do not pretend to be absolutely exact, but the authors claim that, since they have been working with a single strain and their results have always been concordant, the figures obtained are sufficiently exact.

Marchoux and Couvy have been unable to find any coxal gland in argas and they believe that the so-called coxal fluid is in reality coelomic fluid which escapes by a process of rapid filtration. This, they believe, plays no part in the transmission of infection to the vertebrate host.

Although numerous dissections have been made, spirochaetes have been found in exceptional cases only in the salivary glands and then only in small numbers in individuals which have been starved for a fortnight. Examination of the excretory ducts of the salivary glands shewed that they contained considerable numbers of spirochaetes including numerous dividing forms. This was found to be the case even in ticks that had been starved for as long as two months. Further examinations shewed that after a meal the parasites had almost entirely disappeared from the excretory canal and that after an interval of from two to five days they were very numerous in the salivary glands, whence they passed to the excretory ducts.

It is said that a meal of non-infected blood causes a fresh multiplication of the parasite in the tick and fresh invasion of the excretory ducts. An infected tick which had become non-infective through starvation is said to have again become infective as a result of a meal of non-infected blood.

Two explanations of the transmission of the infection through the egg of the tick, which the authors claim to have proved, are given. The secretion of the cephalic glands which serves to cement the eggs together contains spirochaetes and these are capable of penetrating the chitinous covering of the egg. The genital organs contain spirochaetes, as do the salivary glands, and these penetrate the ova. By means of a fine pipette the contents of washed eggs have been withdrawn and long slender spirochaetes have been detected with the ultramicroscope. The contents of 20 eggs injected into a padder failed to infect, but conferred immunity. A fowl's egg which had been incubated for four days was inoculated with the contents of two eggs. On the ninth day after inoculation an emulsion was made of the tissues of the embryo and though spirochaetes could not be detected by direct examination the injection of this material into a padder conferred immunity. The conclusions arrived at are as follows:—

1. The injection of a constant quantity of material obtained by grinding up infected ticks sometimes causes infection and sometimes confers immunity.

2. The infectivity depends upon the number of spirochaetes contained in the ticks.

3. The infective, immunising, and non-infective doses can be measured.

4. All the organs are invaded by the parasite.

5. The spirochaetes pass from the coelom to the acini of the salivary glands and collect in the excretory canal.
6. The salivary secretion is the vehicle of infection.
7. The spirochaetes are present in large numbers in the cephalic glands and are able to penetrate the chitinous membrane of the eggs.
8. The eggs of *Argas persicus* contain spirochaetes, and a single one may contain more than 30.

- (50) LAUNOY (L.) & LÉVY-BRUHL (M.). **L'Infection Spirillaire chez les Poules éthyroïdées. Pouvoir Vaccinant de leur Serum.** [On Spirillar Infection in Fowls deprived of the Thyroid Glands, and the Immunising Power of their Serum.]—*Compt. Rend. Soc. Biol.* 1913. Nov. 14. Vol. 75. No. 31. pp. 352-354.

The authors' conclusions are that in adult fowls, which have been infected with *S. gallinarum*, the thyroid glands appear to play no part in the resistance offered to infection nor in the rapidity with which and the extent to which antibodies are formed.

- (51) LAUNOY (L.). **Le Fer du Sang chez la Poule normale et dans l'Infection par le *Spirochaeta gallinarum* Marchoux et Salimbeni.** [The Iron-Content of the Blood in Normal Fowls and in Fowls infected with *S. gallinarum*.]—*Compt. Rend. Soc. Biol.* 1913. Oct. 24. Vol. 75. No. 29. pp. 248-249.

The iron-content of the blood of the fowl is on an average 0.045 per 100 cubic centimetres.

Bleeding does not exert any influence on the blood content.

In blood taken five days after infection with *S. gallinarum* the iron-content is 25 per cent. lower than the average.

- (52) LAUNOY (L.) & LÉVY-BRUHL (M.). **Sur l'Anémie observée chez la Poule au Cours de l'Infection par le *Spirochaeta gallinarum*.** [On Anaemia in Fowls in the course of Infection with *S. gallinarum*.]—*Compt. Rend. Soc. Biol.* 1913. Oct. 24. Vol. 75. No. 29. pp. 250-252.

The authors' observations have been made upon the blood of fowls inoculated with an attenuated strain of the spirochaetes; parasites appearing in the blood on the second day, and disappearing on the 5th or 6th.

In normal blood the number of red corpuscles varies from 2,180,000 to 3,160,000. During the course of infection a diminution was observed from the time parasites appeared in the blood and the number steadily fell, until about the 5th day it was reduced by half.

By the 12th or 13th day the number had increased again to the normal.

During the course of the infection very small and very large corpuscles were encountered. In addition to these there were many corpuscles which stained faintly or not at all with eosin. The latter was observed from the 3rd to the 10th day.

- (53) HATA (S.). **A Contribution to our Knowledge of the Cultivation of *Spirochaeta recurrentis* [recurrentis].** (Paper read before the Section of Tropical Med. XVII International Medical Congress, London, 1913.)—*Centralbl. f. Bakt.* I. Abt., Orig. 1913. Nov. 26. Vol. 72. No. 1/2. pp. 107-112. With 1 text-fig.

The author has found that NOGUCHI's medium for the cultivation of this spirochaete can be modified in such a way as to obviate the necessity of having ascitic fluid and rabbit kidney. He also finds that it is unnecessary to place sterile paraffin upon the top of the medium.

The ascitic fluid can be replaced by horse serum. The serum is mixed with twice its volume of sterile physiological salt solution and the tubes are placed in a water bath at 58° C., the temperature being very slowly raised until in about three hours it stands at 70° C. The tubes are kept at this temperature for 30 minutes. They are then set aside to cool. The medium forms an opaque semi-coagulated mass.

Instead of rabbit kidney the author uses fragments of the "buffy coat" from the coagulum of the horse serum. This is cut into pieces about 1 centimetre in diameter and two or three pieces are pushed down to the bottom of the coagulated medium.

When this medium is inoculated with blood containing the spirochaete and the tubes incubated to 37° C., multiplication occurs by the second day and actively motile spirochaetes can be found in the medium up to the 10th to the 14th day. The virulence of the organism is maintained in this medium better than in NOGUCHI's. Mice inoculated with a 21 days culture have become infected.

The author has a strain which has been passed through 27 passages in 150 days.

Spirochaetes cultivated at 37° C. will remain alive for about three weeks, but cultures which are kept at room temperature from the 4th or 5th day will retain their vitality for as long as 60 days and possibly longer.

LEISHMANIASIS.

- (54) JANNOT (A.). **Infection de la Souris avec le Virus de la Leishmaniose Canine Naturelle.** [Infection of Mice with Leishmania from Naturally Infected Dogs.]—*Bull. Soc. Path. Exot.* 1913. Dec. Vol. 6. No. 10. pp. 683-685.

The author has succeeded in inoculating two mice with material taken from the bone marrow of two dogs which were found to be naturally infected in Tunis. From these a number of mice, rats, and gerbils—14 in all—were inoculated, but only two mice became infected. Further passage experiments were not carried out.

- (55) LÉMAIRE (G.), SERGENT (E.), LHÉRITIER (A.). **Recherches sur la Leishmaniose du Chien d'Alger.** [Researches on Canine Leishmaniasis in Algiers.]—*Bull. Soc. Path. Exot.* 1913. Oct. Vol. 6. No. 8. pp. 579-581.

In August 1912 the authors took up the examination of the dogs killed in Algiers with the object of ascertaining the percentage infected with leishmania. They were unable to carry on their investigation

continuously throughout the year, but they obtained complete records for the two periods August to November 1912, and April to July 1913.

The results are given in two tables (which appear to contain some arithmetical inaccuracies) and from these it may be gathered that during August and November no infected dogs were found among 44 examined, while during September and October one infected animal was found in each month out of 73 and 88 examined. In this series of animals the examinations involved the preparation of smears from the spleen and bone marrow, four smears being made from each animal.

During the second period of four months, in addition to the preparation of films, cultures were made from the bone marrow.

During this period 272 dogs were examined and seven infected animals were discovered.

It would appear from this that, while the disease can be discovered at all periods of the year, the figures indicate that more cases occur during the months of April and May than during July and August.

It is said that the greater number of cases found during this period is partly due to the fact that a greater number of dogs were examined and to the fact that cultivation was resorted to. In two cases positive cultivations were made when the first examinations of the films had been negative.

A clinical fact noted by the authors is that while some of the infected dogs shew lesions involving the skin, the eyes and the mucous membranes, others shew not the slightest evidence of skin lesions, but only extreme emaciation.

- (56) LAVERAN (A.). *Kala-azar méditerranéen et Kala-azar indien.*—*Bull. Soc. Path. Exot.* 1913. Oct. Vol. 6. No. 8. pp. 574-579.

This paper is a resumé of the report presented by LAVERAN and NICOLLE at the XVIIth International Congress of Medicine held in London in August 1913.

MALARIAL AND OTHER PROTOZOAL PARASITES OF THE BLOOD.

- (57) BRUCE (David), HARVEY (D.), HAMERTON (A. E.), & Lady BRUCE. *Plasmodium cephalophi*, sp. nov.—*Proc. Roy. Soc.* 1913. Oct. 1. Series B. Vol. 87. No. B. 592. pp. 45-47. With 2 plates.

This parasite has been observed in the blood of two duikers in captivity. In one of them the attack was acute and the parasite was present in the blood for four days only. In the other the infection took a chronic course, organisms having persisted in the blood for a period of some months.

The gametes are circular, and the schizonts give rise to 8 to 12 merozoites. Invaded corpuscles shewed marked enlargement and paleness of colour. The pigment is collected into a single mass. There is intense concentration of the portion of the red cell within the food vacuole. The parasite is remarkable for its size, free merozoites sometimes measuring 3.5 by 4 microns, this being about the size of the normal red cell of the antelope. A full-grown schizont before cleavage of the protoplasm may measure 10 microns in diameter.

The duiker in which the infection ran a chronic course was captured

near the lake shore, and parasites were found in its blood a few weeks after its arrival at Kasu Hill. The animal was about three months old. It was probably infected before capture. The other animal was captured on the plateau and had been in captivity for six months before being brought to Kasu. It would appear that its infection was contracted locally.

The infection was discovered at the height of the dry season and though a systematic search was made no mosquitoes could be found.

Another young duiker, a reedbuck and a hartebeeste were in the same compound, but no parasites could be discovered in their blood.

- (58) LEGER (M.) & BOUILLIEZ (M.). *Recherches Expérimentales sur Plasmodium inui Halberstaedter et Prowazek d'un Macacus cynomolgus*. [Experimental Investigations regarding *Plasmodium inui* of *Macacus cynomolgus*.]—*Ann. Inst. Pasteur*. 1913. Nov. Vol. 27. No. 11. pp. 955-985.

This paper is an amplification of those already published by the authors (see this *Bulletin* Vol. 1, Abstracts Nos. 172 and 531). The points dealt with are the following:—morphology, transmission by inoculation, the course of the experimental infection, attempts to transmit the infection to animals other than monkeys, the effects of quinine and splenectomy, attempts to cultivate the parasite, and the identification of the organism.

In the fresh state the parasites appear as round, ovoid, or amoeboid bodies within the corpuscles. Young parasites are difficult to recognise as they do not contain any pigment, and the gametes are distinguishable from the schizonts by the fact that the pigment contained in them is greater in amount and the individual granules are of larger size. The only free parasites seen are the gametes and young merozoites. The authors have never observed flagellation of the microgametocytes.

In stained preparations the following details can be observed. Young schizonts are round in shape and measure rather less than 2 microns in diameter. They are devoid of pigment and their vacuole-like nucleus contains a caryosome. This caryosome is sometimes rounded in shape and centrally placed, but it may be peripherally placed, producing a signet ring appearance. Occasionally two caryosomes are present, and these may be placed at opposite poles of the parasite, or side by side and connected by a delicate filament. A single corpuscle may be occupied by two or three parasites.

In older parasites the caryosome may be curved or rod-like. As the schizont increases in size, it becomes rounded and its protoplasm contains an increasing number of small pigment granules.

The maximum number of merozoites produced by schizogony is 16, and the pigment becomes collected together into a small mass in the centre of the parasite.

Microgametes are rounded or polyhedral, and contain a number of pigment granules scattered through their body. The nucleus, which occupies about a quarter of the parasite, takes the form of a number of rods of chromatin embedded in a pink matrix. Macrogametes are usually larger than the male elements and are often somewhat square in shape. Their protoplasm stains a deep blue tint. The pigment granules are smaller than in the microgametocyte and the nucleus stains deeply.

Red corpuscles containing parasites are not enlarged nor deformed, and they stain as intensely as those containing no parasites. At an advanced stage of infection there is marked anisocytosis with a preponderance of megalocytes, and it is remarkable that these are practically never invaded by parasites, while the microcytes frequently are. Polychromasia is not uncommonly seen, but no punctate basophilia has been observed.

The schizogonic cycle occupies 48 hours.

The parasite does not correspond exactly with any of the three malarial organisms of man, but possesses to some degree the principal characters of all three.

Inoculating intramuscularly, subcutaneously, and intraperitoneally, the authors have passed the virus through seventeen passages, and have infected the following species:—*Macacus cynomolgus*, *M. sinicus*, *M. rhesus*, *M. nemestrinus*, *Cercopithecus callitrichus*, *C. patas*, *C. cephus*, and *Papio anubis*. Two chimpanzees, a *Cercocebus fuliginosus*, and a maki were found to be refractory.

The disease as observed in animals experimentally infected occurs in two forms—acute and chronic.

The acute form is the more frequently seen. Parasites make their appearance in the blood in from 4 to 9 days after inoculation, and death takes place in from 10 to 18 days, the period elapsing between the appearance of parasites in the blood and death never exceeding 10 days. Fifty per cent. of the red corpuscles may be invaded and a single corpuscle may contain three parasites.

At the post-mortem examination there is not always observed enlargement of the liver and spleen, but these organs are grey or even black in colour. Preparations from these organs and from the lungs in some cases shew that there is an abundant infiltration with pigment. This is sometimes free and sometimes included within large mononuclear leucocytes and epithelial cells.

The virulence of the organism was neither exalted nor attenuated by the passages.

In the chronic form of the disease the period of incubation is not sensibly longer than in the acute form, and it is characteristic of this form of the disease that there are periods during which the parasite disappears from the circulation. After their first appearance in the blood they steadily increase in numbers for a time and then disappear either suddenly or gradually. The crisis may last for a week or even a month. During the relapse which follows parasites may be as numerous or slightly less numerous than during the crisis. As a rule there are two relapses and exceptionally three.

During a period of five or six weeks the animal does not appear to suffer in health, but subsequently it becomes dull and ceases to feed. Contrary to what is observed in the acute cases there is sometimes diarrhoea, but the haematuria and haemoglobinuria observed in acute cases have not been seen in chronic ones. The infection causes little or no rise of temperature.

All the monkeys in which the disease took a chronic course, with the exception of one, died after a longer or shorter interval. At the time of death there were no parasites in the blood and none had been observed for a variable length of time previously. Although parasites could not be discovered, the blood of one monkey which survived

223 days proved infective by inoculation. One animal had survived for nine months at the time of writing. The authors have failed to infect animals other than monkeys. Quinine as a prophylactic was found to be of doubtful value, and as a curative to be useless.

Splenectomy was practised upon an animal six months after inoculation when it appeared to have been free from parasites for some time. Parasites again made their appearance in the blood after the operation and the animal died during the following month. The operation was performed upon a second monkey before inoculation and the animal died of the chronic form of the disease four months afterwards.

The authors never observed the morphological changes in the parasite described by BUONIOI as occurring in animals upon which splenectomy had been performed.

Following BASS's technique the authors have succeeded in obtaining a continuation of the development of the parasite in vitro at about 22° C., but subcultures failed in every case. Citrated infective blood kept upon ice proved to be infective five days later and in another instance well stained preparations could be made from blood preserved in a similar manner for a month, but an animal inoculated on the eighth day failed to become infected.

The authors believe that the parasite is *Plasmodium inui* Halberstädter and Prowazek, to which species they also assign *Pl. cynomolgi* Mayer.

- (59) JOYEUX (C.). Note sur quelques Protozoaires Sanguicoles et Intestinaux observés en Guinée française. [Some Protozoal Parasites of the Blood and Intestines observed in French Guinea.]—*Bull. Soc. Path. Exot.* 1913. Nov. Vol. 6. No. 9. pp. 612-615.

No new parasites are described in this paper, the author's object being to add to knowledge regarding the distribution and frequency of organisms already known.

- (60) VISENTINI (A.). Gli Emoparassiti della Talpa in Italia. [The Blood Parasites of the Mole in Italy.]—*Arch. f. Protistenkunde.* 1913. Dec. 11. Vol. 32. No. 2. pp. 257-266. With 1 plate.

In 30 per cent. of the moles examined the author has found *Trypanosoma talpae*, and *Elleipsosoma thomsoni* in 4.34 per cent. He has also observed *Grahamella talpae* Brumpt, regarding the parasitic nature of which there appears to be great uncertainty.

- (61) MOLDOVAN (J.). Beitrag zur Entwicklung des *Leucocytozoon ziemanni* (Laveran). [The Development of *Leucocytozoon ziemanni*.]—*Centralb. f. Bakt.* 1. Abt. Orig. 1913. Sept. 27. Vol. 71. No. 1. pp. 66-69. With 1 plate.

Two screech owls, which had shewn the gametes of this parasite in their blood during life, were examined by the author after death, and in films prepared from the lungs, heart, brain, liver, kidneys and bone marrow young endocellular schizonts were found. In addition to these, young male and female gametes were found. The schizonts contained as many as 30 nuclei. The spleen was not examined.

The young schizonts shew a central nucleus of loose texture surrounded in most instances by particles of chromatin. The cytoplasm is alveolar in structure.

As the parasite increases in size changes are to be observed in the nucleus. This becomes divided into a larger loose-textured portion and a smaller portion of very close texture. The smaller portion takes part in the formation of the nuclei of the schizonts, while the larger portion gradually increases in size, becomes less distinctly marked off, and finally disappears.

The segregation of the cytoplasm around the nuclei and the separation of the merozoites has not as yet been observed.

In a small proportion of the parasites nuclear division does not occur.

TOXOPLASMOSIS.

- (62) PIXELL (H. L. M.). *Notes on Toxoplasma gondii*—*Proc. Roy. Soc.* 1913. Oct. Series B. Vol. 87. No. B. 592. pp. 67-77. With 1 plate.

The parasite proves fatal to mice inoculated intraperitoneally in from five to 15 days. There may be an abundant or scanty peritoneal exudate, and the number of parasites present in this is not in proportion to the amount of exudate. Often parasites are scantily present in cases in which the infection runs an acute course. The parasites in the exudate are either free or enclosed in cells—mononuclear leucocytes or macrophages. Examination of the mesentery shews that the endothelial cells and the connective tissue cells may contain large numbers of parasites. The abdominal and thoracic organs may contain a few.

Multiplication takes place by a process of simple division. Occasionally a number of parasites situated side by side divide in different planes producing an appearance suggestive of cyst formation. Attempts to cultivate the parasite on a number of different media failed.

The author was unable to infect pigeons from mice, and it is stated that NICOLLE has observed the same fact.

- (63) CARINI (A) & MACIEL (J.). *Toxoplasmose naturelle du Chien*. [Natural Toxoplasmosis of the dog.]—*Bull. Soc. Path. Exot.* 1913. Dec. Vol. 6. No. 10. pp. 681-683.

Two puppies which were born in the Institute (S. Paulo, Brazil) had been inoculated with the virus of a little-known disease of the dog termed "nambiuvu." The animals died within a fortnight of each other (after an interval which is not stated) and, while the parasite of "nambiuvu" could not be detected, *Toxoplasma* was found.

Post-mortem examinations were made immediately after death and the following lesions were found:—a small amount of serous exudate in the thoracic and abdominal cavities, some areas of pneumonia in the lungs, and marked hyperaemia of the liver and kidneys. *Toxoplasma* was especially numerous in the lungs, but it was also found in the liver, kidneys, bone marrow, and spleen.

An emulsion of the organs of each of these dogs was used for the successful inoculation of other dogs and pigeons.

Young dogs are particularly susceptible, and death takes place in from 8 to 12 days, the lesions found at the post-mortem being identical with those encountered in natural cases. Adult dogs as a rule fail to become infected even when massive doses of virus are used.

The pigeons died in from 12 to 19 days. The parasite has been passed through a series of pigeons, and in some instances a symptom which has already been recorded by one of the authors, namely oedema of the eyelids and cloaca, has been observed. The parasite is very numerous present in the oedematous tissue.

In view of the facts that the morphology of the parasite occurring in natural and experimental cases in the dog and pigeon is identical with that found in the rabbit and pigeon, and the pathogenic ranges of *Tox. canis* and *Tox. cuniculi* are the same, it appears to be very doubtful whether the two should be considered to be different species.

HELMINTHS.

- (64) LEIPER (R. T.). *Seven Helminthological Notes.*—*Jl. London School Trop. Med.* 1913. Nov. Vol. 2. Part. 3. pp. 175-178. With 1 text-fig. [(d.) Flukes in the Dog in Asia. pp. 176-177.]

The trematodes recorded hitherto from the dog in Asia are: *Schistosoma japonicum*, *Paragonimus westermanii*, *Clonorchis endemicus*, and *Opisthorchis neverca*. As a result of the examination of material sent from Formosa three further parasites have to be added to the list. These are:—*Heterophyes heterophyes*, *Yokogawa yokogawa*, and a new species *Centrocestus cuspidatus*, var. *caninus*.

With the exception of the last these flukes are parasitic in man. *Heterophyes* has been found in two instances, in a Chinaman and in a Japanese at the Albert Dock Hospital. *Yokogawa* is said to be sporadic in man and the dog in Korea, Japan, and Formosa. A single specimen of *Centrocestus* was present in a slide of faeces from the dog and, although the parasite closely agrees with that described by Looss as occurring in the kite in Egypt, Leiper follows Looss's ruling and classifies the parasite of the dog as a distinct variety naming it *Centrocestus cuspidatus*, var. *caninus*.

The specimen from the dog differs from that described by Looss in that there are only 28 spines round the mouth instead of 36, but some of the spines may have been lost.

- (65) PRICOLO (A.). *Sur la Filaire hématique du Chameau.* [On the Blood Filaria of the Camel.]—*Centralbl. f. Bakt.* 1. Abt., Orig. 1913. Oct. 4. Vol. 71. No. 2/3. pp. 199-200.

In this paper the author describes the adult forms of the larval filariae found by him in the blood of camels in Tripoli and described in the *Centralblatt für Bakteriologie* 1913, January 11.

He states that he has placed beyond all doubt the relationship between the larvae and the adults.

Filariasis of the camel generally occurs during the hot weather, and the larvae are almost exclusively found in the blood during the night.

Male and female parasites have been found in the blood vessels of

the lung upon two occasions, and in those of the testicles on six. They appear to be capable of escaping easily from the lumen and, except during the first stage of their existence, they do not appear to become attached to the vessel walls.

The lungs in which the author found the parasites shewed calcareous nodules, but the association between these and the worms is doubtful.

In infested testicles there is a condition of hypertrophic sclerosis. The testicles are increased in size and are firmer than normal, the surface presenting a bosselated appearance. Microscopic examination shews that the testicular substance is for the most part replaced by connective tissue.

The female has the appearance of a thick strand of catgut with a slight tinge of pink colouration. The length of the worm is about 22 centimetres and the thickness 666 microns. The head is large, measuring about 140 microns, and it is suddenly constricted posteriorly to form the neck, which is about 117 microns in length and rather more slender than the head. The body of the worm attains its maximum thickness at about the point where the ovaries first become visible. The tail terminates in a truncated point.

The ovaries contain thousands of cells in different stages of development. They are polygonal in outline, measuring about 30 microns in length and varying from 11 to 20 in width.

The eggs contain embryos in different stages of development and measure 32 by 12 to 15 microns.

The male worm resembles a thin strand of catgut and measures from 8 to 12 centimetres in length by 300 microns in thickness. The head, which measures 105 microns, is separated by a very short and slightly marked constriction from the body. The caudal extremity of the body is very slender and is shaped like a corkscrew with three turns.

The presence of the adult worms in the testicles does not appear to give rise to any symptoms. The pathogenic rôle of the larvae has not yet been investigated.

The author suggests the name *Filaria haematica cameli* for the parasite.

- (66) PRICOLO (A.) *Strongle capillaire du Chameau*. [Strongylus of the Camel.]—*Centralbl. f. Bakt.* 1. Abt., Orig. 1913. Oct. 4. Vol. 71. No. 2/3. pp. 201-202.

The author has found this parasite in the small intestine of camels in Tripoli.

The males and females are about the same size with the exception that the posterior portion of the male is a little more slender than the corresponding portion of the female.

The worm measures from 5 to 7 millimetres in length and it gradually increases in thickness from the head, which measures about 18 microns, to a maximum of 100 microns, which it attains close to the posterior extremity of the body. The female ends in a short cone. Occasionally there is a small appendage resembling a wing on either side of the head.

The head is curled round in a hook-like manner, but this is probably a post-mortem change. The mouth is unarmed. The uterus appears

to be straight and single, and the eggs which are double-contoured measure 90 by 45 microns.

The anterior extremity of the male worm is not hook-like. At a point 10 microns in front of the bursa is situated the anus which has a brown colour and is shaped like a fork with three prongs. The posterior prong is slender and cylindrical and is directed towards the bursa and terminates in a kind of knob. The two posterior (anterior?) prongs are wide and flattened, and are directed forwards enclosing a space which is divided into two by two small appendages attached to the anterior prongs.

The worm appears to be of very constant occurrence in the small intestine of the camel, as the author has found it whenever he has made a search. It is frequently associated with haemorrhages, hyperaemia and catarrh of the mucous membrane.

Possibly the worm is that which has been described under the name *Strongylus instabilis* in other mammals.

ANTHRAX.

- (67) HOLMES (J. D. E.). **A Note on the McFadyean Staining Reaction for Anthrax Bacilli.**—*Agricultural Research Institute, Pusa, Bulletin* No. 36. 1913. pp. 1-3. With 1 plate. (Superintendent Government Printing, Calcutta.)

The author shews that the view expressed by MITTER (*Jl. Trop. Vet. Sci.* Vol. 4, No. 1, pp. 68-70) regarding a supposed difference between the staining reactions of anthrax bacilli in Bengal and in Europe is incorrect, the probability being that the technique of the McFadyean method of staining was incorrectly carried out.

- (68) HOWARD (G. G.). **Anthrax in Elephants.**—*Vet. Record.* 1913. Aug. 2. Vol. 26. No. 1308. pp. 69-71.

This note describes briefly outbreaks in two herds and the author explains that the brief nature of the record is due to the difficulties in the way of obtaining a more detailed one.

The animals involved in the outbreak lived entirely in the jungle at distances varying from 2 to 12 days march from headquarters. During the "off" season (March, April, and May) they are caught once daily and brought into camp for inspection by the head rider and then released again to graze.

At the commencement of the outbreak three animals died and the diagnosis was made upon the examination of blood smears forwarded to headquarters. After forced marching the author arrived at the infected area on the following day and found another animal shewing signs of illness. The animal was dull and had a haggard look. There was a swelling on the right side just below the scapula which was hot and painful. Examination of the blood while the animal was still alive shewed that a few anthrax bacilli were present in it. The animal died early the following morning, six hours after the bacilli had been found in the blood, and at the time of death bacilli were very numerous present.

The same day another animal was observed to be dull, and to have

a watery discharge from the eyes. In this case also there was a small well-defined swelling below the right scapula resulting in lameness. In both of these animals it was impossible to get any temperature taken on account of their viciousness. The swelling was continuously fomented with hot water and on the fourth day it burst in three places. Bacilli were not found in this animal's blood, and although the animal lost greatly in condition it eventually recovered. On the day that this animal was first observed to be ill another elephant shewed symptoms and it died on the following day, anthrax being confirmed by microscopic examination. Three days later another animal died of anthrax and one other became ill but recovered. In four of the animals there was a swelling on the right side.

RABIES.

- (69) NOGUCHI (H.). **Contribution to the Cultivation of the Parasite of Rabies.**—*Jl. Experimental Med.* 1913. Sept. Vol. 18. No. 3. pp. 314-316. With 1 plate.

This paper contains a brief account of what the author claims to be the parasite of rabies in artificial cultures. The method employed was that used for the cultivation of the spirochaete of relapsing fever.

In cultures made either from the "street," "passage," or "fixed" virus minute granular and coarser pleomorphic chromatoid bodies make their appearance and these can be found in sub-cultures through many generations. The smallest of these bodies are just on the borders of visibility with a 2 mm. lens. On a number of occasions the author has observed in certain cultures rounded or oval nucleated structures provided with an enclosing membrane and quite different in appearance from the granular bodies. The appearance of these bodies in the cultures was sudden and after persisting for four or five days they disappeared, their disappearance coinciding with an increase in the number of small granular bodies.

Experimental inoculation of dogs, rabbits, and guinea-pigs with cultures containing the nucleated or the granular bodies has given rise to rabies, as shewn by the typical symptoms and positive animal inoculations, the granular and nucleated bodies being found in the brains of the animals so infected.

A paper substantially the same as this appears in the *Revue Vétérinaire*, 1913. Oct. Vol. 30, No. 10, pp. 592-595.

- (70) LEVADITI (C.). **Virus Rabique et Culture des Cellules "in vitro."**
[The Virus of Rabies and the Cultivation of the Cellules in vitro.]
—*Compt. Rend. Soc. Biol.* 1913. Dec. 12. Vol. 75. No. 35. p. 505.

In this preliminary note the author states that using the technique of BURROWS, HARRISON and CARREL he has succeeded in cultivating spinal ganglia of rabid monkeys in monkey plasma. The pieces have been transferred to fresh plasma regularly every five or six days. It has been found that the virus of rabies preserves its virulence for the rabbit for at least a month at 37° C. Rabbits inoculated at each passage with portions of the cultivated ganglia have contracted rabies.

The two rabbits inoculated into the brain with the virus of the 5th passage and after thirty days cultivation became rabid on the 10th and 11th days.

Full details regarding the technique and the results of microscopic examination will be given in a future paper.

- (71) LUZZANI (Mme L. N.). **Le Diagnostic de la Rage par la Démonstration du Parasite Spécifique. Resultats de Dix Ans d'Expériences. (Première Partie.)** [The Diagnosis of Rabies by the Demonstration of the Specific Parasite (First Part.)]—*Ann. Inst. Pasteur*. 1913. Nov. Vol. 27. No. 11. pp. 907-924.

This article contains the author's views regarding the parasitic nature of Negri bodies and a brief description of a process of spore formation in which the central nucleus divides into a number of pieces, each of which has separated off round it a portion of the general body substance. The bulk of the paper is made up of a summary of the reports and statistics published by a large number of authors and a repetition of the various methods of staining that have been devised for the demonstration of Negri bodies. It is stated that while in cases of furious rabies the bodies are to be found in Ammon's horn, the cerebral cortex and cerebellum, in cases of dumb rabies they may be absent from these situations but present in the spinal ganglia and spinal cord.

- (72) LUZZANI (Mme L. N.). **Le Diagnostic de la Rage par la Démonstration du Parasite Spécifique. (Deuxième Partie.)** [The Diagnosis of Rabies by the Demonstration of the Specific Parasite. (Second Part.)]—*Ann. Inst. Pasteur*. 1913. Dec. Vol. 27. No. 12. pp. 1039-1062.

The author advises the adoption of the following procedure for the detection of Negri bodies in suspected material. In any case the hippocampus should first be examined if that be available. Transverse sections are cut from the fresh specimen and scrapings are taken from the cut surface. These are examined in the moist state without staining by spreading them out on a slide with a little dilute acetic acid. A plate is given shewing the appearance presented by the nerve cells containing the bodies.

Should this method fail, a thin slice of the hippocampus is fixed in Zenker's fluid for four hours and then washed in water, after which scrapings are taken from the cut surface and treated in the same manner as the fresh specimens. In the event of a negative result the thin slices are rapidly dehydrated and passed through chloroform and embedded in paraffin. The sections are stained by Mann's method.

Reference is then made to cell inclusions other than Negri bodies and the literature concerning them is discussed at some length. The characters of those found in cases of distemper are contrasted with those found in rabies.

The author draws attention to the discovery in the cells of the hippocampus and of the cerebellum of small cell inclusions in the brains of healthy cats. These stain red with Mann's method, but they have not the typical structure of Negri bodies.

The paper includes tabulated results of examinations made by the author.

- (73) BERTARELLI (E.) & MELLI (C.). **Experimentelle Untersuchungen über die Pseudolyssa.** [Experimental Investigations regarding False Rabies.]—*Centrabl. f. Bakt.* 1. Abt., Orig. 1913. Oct. 11. Vol. 71. No. 4. pp. 286-291.

The following points were investigated by the authors :—

1. The filtrability of the virus.
2. The infectivity of the peripheral nerves.
3. Whether the virus can pass along the course of nerves.
4. Whether cell inclusions comparable to those found in rabies occur in the nerve cells of the central nervous system and spinal ganglia.

The only one of these points which has already received attention at the hands of experimenters is the first.

The virus used was obtained from CARINI.

In the first filtration experiment the brain of a rabbit infected by subcutaneous inoculation was mixed with 10 cc. of physiological salt solution and filtered at a pressure of one atmosphere through a new Berkefeld filter which had been tested with *B. prodigiosus*. Two rabbits inoculated respectively subcutaneously and subdurally with the filtrate failed to become infected.

In the second filtration experiment the "virus" was macerated for several hours in salt solution and then greatly diluted (no details given). This liquid was filtered at $\frac{3}{4}$ atmosphere pressure. The filtrate was centrifuged and used to inoculate one rabbit intraperitoneally and another subdurally. In both cases the result was negative.

A portion of the sciatic nerve of a rabbit which had died three days after subcutaneous inoculation was ground up in a mortar with salt solution, the liquid being used to inoculate rabbits subcutaneously and intraperitoneally. Neither of the rabbits died.

Another rabbit was inoculated with a similar liquid prepared from the sciatic nerve of a rabbit which had died as a result of inoculation into the sciatic nerve of the opposite side. The result was again negative.

The experiment was repeated with the sciatic nerve taken from a dog which had died as the result of subcutaneous inoculation from a rabbit. In this case also the result was negative. From these experiments the conclusion is drawn that the peripheral nerves are non-infective, this being in marked contrast to what is the case in rabies.

To ascertain whether the virus traverses the course of the nerves an animal (species unstated) was inoculated into the sciatic nerve which had been exposed and isolated with pads of wool. Death occurred with typical symptoms two days later.

As soon after death as possible the inoculated nerve was removed, ground up with salt solution, and used for the subcutaneous inoculation of two rabbits. Both died of the disease. The brain of one of these rabbits proved infective when inoculated into another animal.

A rabbit inoculated into the sciatic nerve which had been ligatured above the seat of inoculation failed to become infected. The same result was obtained when a rabbit was inoculated from a portion of

the sciatic nerve cut out above the seat of inoculation immediately after the inoculation had been performed.

Infection followed the inoculation of a very small quantity of virus into the muscle of the gluteal region which was exposed for the inoculation in order to avoid injuring any nerve trunk.

The conclusion that naturally follows from these results is that the virus can become disseminated not only along the course of nerves but in some other manner also. Some of the experiments suggest, however, that the dissemination of the virus along the nerve trunks is specific as is the case in rabies. It is said to be possible that the dissemination of small quantities may take place through other channels.

The authors have examined the brain of only one dog with the object of ascertaining whether bodies resembling Negri bodies are present, and only a single body in any way resembling a Negri body was found.

RINDERPEST.

(74) BRADDON (W. L.). **Some Peculiar and Probably Specific Bodies in the Erythrocytes in Rinderpest and another Allied Disease.**—*Parasitology*. 1913. Oct. Vol. 6. No. 3. pp. 265-275. With 1 plate.

The author's observations were made in connection with the production of a protective serum for buffaloes in the Malay States.

For the detection of the bodies described a special method of treating the blood is essential, the method being one which the author has been in the habit of using for blood-studies.

Method. The blood is diluted with a mixture containing 1 per cent. of citrate of potash and 0.5 per cent. of ordinary medicinal methylene blue. Different proportions of the fluid must be added varying from one-fifth to one-third of the volume of blood, according to the condition of the latter, which as taken from the vein may be almost tarry or excessively hydraemic.

"Of the mixture, some specimens are put up under cover-glasses, sealed at the edges with a paraffin of low melting-point; other portions are put in watch glasses exposed to the air, but not to light, and are kept in Petri dishes arranged as moist chambers. When examination reveals the presence of the specific bodies in these preparations, films may be spread in the ordinary way, and fixed as permanent specimens."

Appearances presented by the bodies. After a period varying from 6 to 48 hours there appears upon the blood corpuscle a blue point, from which there later develop one or more delicate fine lines going deeper into the corpuscle. These lines by extending and thickening usually demarcate a long slender needle- or sword-shaped body which may extend into, across, or through the corpuscle.

The bodies may be straight or curved, may present a single or double contour and may vary considerably in width. The ends may be pointed or oval. In some cases the protruding ends of the bodies carry the membrane of the corpuscle with them. The bodies may appear to be in one plane, bent, twisted, or folded. A single cell may contain one or more such bodies.

The bodies may be completely extruded from the corpuscles, and many filamentous and lanceolate forms may be seen either attached by one end to the corpuscle or free in the plasma.

In specimens taken from acute cases when near the crisis none of these forms may be found, but corpuscles shewing a minute speckling with blue points may be seen. In a small number of corpuscles fewer granules of a larger size may be observed, and in some of these the granules may have a comma-like form.

The presence of the endoglobular bodies is indicated in the author's opinion in other ways. The blood corpuscles undergo an alteration of outline, appearing as though a slice of variable size had been cut off from one side. Measurement is said to shew that there is in reality no loss of substance, but that the alteration of shape is due to some pressure exerted by a body within the corpuscle. The author has been unable to demonstrate this body within the corpuscle by staining.

Movements. Within the first 24 hours of observation corpuscles and particularly those which appear to have had a portion sliced from them shew active movement. Rapid oscillations lasting for a short time are combined with rotatory changes in position. The effect is an exaggerated Brownian movement, and the corpuscles actively jostle one another.

Within the corpuscles, even when in a fairly advanced stage of staining and after several days of observation, the bodies shew movements of their own. They bend and unbend, twist or rotate sluggishly on their long axis, change position within the corpuscles; when attached by one extremity to the exterior of the corpuscle they sway freely in the plasma and vibrate with a limited range. The free forms move through the plasma shewing movements of rotation, bending and unbending.

Relation to disease. The bodies have been found invariably in all cases of acute rinderpest during the febrile stages, and in the great majority of cases up to eight months after recovery.

In artificially infected animals which have first been proved to be free of these forms they appear as early as the third day after inoculation. From this date they increase rapidly in numbers, and even in serum-protected animals three-quarters of the corpuscles may become invaded. Their appearance precedes the onset of pyrexia by several days and thus forms a valuable means of detecting contacts.

In animals less susceptible than buffaloes the bodies are smaller, less numerous, and less active. In successive reactions following the injection of virus into immunised animals there is also a progressive diminution in number and size.

In one of two domestic pigs inoculated numerous slender forms were found in the blood on the fourth day. In goats which act as passive carriers of the virus (which in them is active from the 4th to the 8th day after inoculation), the bodies could not be distinguished in any of the shapes occurring in cattle, but tapering filaments were observed to be protruding from opposite sides of the corpuscles, and these were observed to continue unaltered in shape for ten days or more.

A New Rinderpest-like Malady. The author's attention was drawn to a disease among buffaloes which resembled rinderpest in its clinical features and in the post-mortem findings, but which occurred as a limited epizootic and caused only a low mortality.

Examination of blood from these cases by the author's method failed to reveal the bodies observed in true rinderpest, but shewed an intracorpuseular body of a different kind.

In nearly every corpuscle a short thick rounded, ovoid, angular, or nearly square body could be seen. They stained of a dark green to a blue black colour and appeared to consist of a vesicle measuring from $\frac{1}{2}$ to 3 microns in diameter distended with stain. They appeared to have a tendency to protrude from the corpuscles but none were found absolutely free.

Remarks as to technique. The only method by which the author has been able to stain more than the mere margins of the bodies is as follows:—The specimen, having had any excess of mercury (sublimite is the best fixative) and blue washed off, is immersed in the original solution for five minutes and overstained. It is then washed and treated for 10 seconds only with a mixture of 2 per cent. orange G., 1 per cent. Anilin blue, and 2 per cent. Oxalic acid in water. Wash quickly with water, absolute alcohol, acid alcohol, and again water. In successful specimens the corpuscles have a green tint and the specific bodies have a deep brown colour.

It appears to be essential that the stain in the first instance should be applied to the unfixed corpuscle.

(75) JOHNSON (J. C.). **Observations on Mammalian Erythrocytes.**—*Parasitology*. 1913. Oct. Vol. 6. No. 3. pp. 276-278.

The observations recorded in this short paper were carried out with the object of ascertaining whether any appearances resembling those described in BRADDON's paper on rinderpest could be discovered in the blood of healthy and diseased animals.

Normal blood was examined from a man, two dogs, two rabbits, a white rat, a calf, and a ram. The diseased animals from which blood was taken for examination included a calf infected with *Theileria parva*, three dogs suffering from acute piroplasmosis, two dogs suffering from chronic piroplasmosis, and a horse "salted" against infection with *Nuttallia equi*.

Several specimens of blood were taken from each animal by running it into marked tubes containing BRADDON's mixture, and in proportions varying from $\frac{1}{6}$ to several times the volume of staining fluid.

With the blood of normal animals it was found that the quantity recommended by BRADDON was insufficient to prevent coagulation.

Four methods of examination of the erythrocytes were adopted:

1. A drop of mixture from the tubes was transferred to a slide, covered, and ringed with vaseline.

2. Coverglass films were made from the mixtures, fixed wet over formalin, or with saturated mercuric chloride, and mounted in balsam.

3. Films of fresh blood were made at the time of collection, and treated with either Giemsa's or Leishman's stain.

4. For correlation with the above, unstained films were observed of the blood mixed with Hayem's solution.

A great many films had to be rejected owing to osmotic changes in the red blood corpuscles.

In specimens in which the mixtures were correctly made, so as to avoid change in the histological characters of the blood corpuscles, the solution recommended by BRADDON was found to be an excellent preservative for blood films. Several ringed films kept in perfect condition for over a month.

After prolonged examination of many films the author concluded that :—

1. The appearances of the erythrocytes in the morbid conditions examined were easily differentiable from those described by BRADDON.

2. Normal blood corpuscles from the above sources presented nothing like his description of the alleged parasite of rinderpest, which tends to support his view that what he describes is specific to that disease.

MISCELLANEOUS.

- (76) MINETT (E. P.). **Report of Epidemiological Survey and Investigation into Probable Causes of Sickness amongst Mules on Plantations Bath, Blairmont, Providence and Springlands, British Guiana.**—*Jl. Trop. Med. & Hyg.* 1913. Dec. 1. Vol. 16. No. 23. pp. 362-364.

The disease which is briefly described in this paper has been the cause of death of a number of horses and mules in the districts mentioned. The period of incubation is uncertain, but probably amounts to several weeks at the most. The symptoms are :—Loss of condition, progressive anaemia, paralysis of the hind quarters and in some cases haemoglobinuria. Jaundice was seldom observed. Acute cases terminate fatally in 3 weeks, while chronic ones may last for 6 months. The lesions found are as follows :—Anaemia of all the organs, haemorrhages under the serous membranes, no enlargement of the spleen. No trypanosomes were found and examination for meningococcus was negative. Sections of the spinal cord shewed a small amount of round-celled infiltration around the capillaries.

In a Supplementary Report on Mule and Horse Disease in the County of Berbice it is stated that smears of blood taken during the febrile period shewed large numbers of trypanosomes "closely resembling *T. equinum*."

- (77) HADWEN (S.). On "Tick Paralysis" in Sheep and Man following Bites of *Dermacentor venustus*. With Notes on the Biology of the Tick.—*Parasitology*. 1913. Oct. Vol. 6. No. 3. pp. 283-297. With 2 plates.

History of the disease. During the spring of the years 1910 to 1913 complaints were received from Keremeos, British Columbia, that a peculiar form of paralysis among sheep had been observed.

In 1911 an outbreak occurred among 900 sheep imported from Montana, and during March and the first week in April 46 old sheep were lost.

From March 1st to 15th 1912, 34 yearling sheep were affected and 10 died. In May, June and July about 300 were affected, and about 80 died. There is no definite information that these were affected in the same way, but it was said that they were suffering from a form of paralysis.

From March 25th to April 5th 1913 five yearlings were paralysed, but none died.

The author proceeded to the district in April, and though he saw no recent cases some which had been affected were shewn to him. The

animals had just been shorn and on examining one of them a large tick which had been cut in half was found attached to its back.

Surrounding the tick was a large discoloured area and this caused the suspicion that the disease was due to the bites of ticks.

Recently papers have been published recording the occurrence of a peculiar form of paralysis occurring in human beings as a result of tick bites.

On returning to Agassiz the author was able to reproduce the disease experimentally with ticks brought from the district where the naturally contracted cases had occurred.

In at least one instance it was found that the removal of a tick from an affected animal was followed by prompt recovery.

The explanation of the occurrence of a few cases only in lambs lies in the fact that by the time they are taken up to the hills the ticks have practically disappeared for the season.

The tick season around Keremeos may begin as early as February and does not extend much beyond May, but the author was informed that occasionally a few ticks were observed on sheep during the autumn and that cases were sometimes encountered during that part of the year.

Nature of the disease and symptoms presented. The disease occurs in sheep and man, and probably in other species also, and is caused by the bite of *Dermacentor venustus*.

The tick has a decided preference for biting along the spinal column, but it remains to be shewn whether the disease is produced by the bites of the tick on any other portion of the body. The tick certainly produces serious local effects wherever it may bite.

The disease is said in some cases to last for months.

Reports have been received regarding the occurrence of a disease of the same or a similar nature in colts, dogs, rabbits and grouse.

In the sheep symptoms of paralysis occur in from 6 to 7 days after the ticks have become attached to them, and the only lesions found at the post-mortem are subcutaneous haemorrhages where the ticks have been attached and congestion of the brain and cord. Attempts to transmit the disease by inoculation have failed and no parasites have been discovered in the blood or nerve tissues.

Broth cultures prepared from diseased animals have remained sterile. The probable explanation of the disease is that it is due to toxins injected by the ticks and that these are elaborated at a time when engorging is rapid, that is 24 hours before the ticks drop off.

The average time occupied by female ticks in becoming engorged was $6\frac{1}{2}$ days.

The period of incubation suggests that the disease may be of microbic origin and it is possible that the causal agent has not been recognised, but the fact that no fever has been recorded and inoculation experiments have proved negative rather contra-indicates this.

The fact that old sheep were attacked during the first year and that only yearlings were subsequently diseased suggests that an attack of the disease confers immunity. This does not interfere with the toxin theory of the disease.

So far there seems to be no evidence connecting the paralysis observed among the sheep with Rocky Mountain fever, the only other disease transmitted by *D. venustus*.

The disease develops somewhat slowly and in cases where recovery is about to take place the first stages towards recovery also are prolonged, after which progress is rapid.

It is suggested that the removal of ticks and dipping would prove of benefit in the treatment of the disease.

The male ticks attach and detach themselves, causing hæmorrhage at every bite. The blood seems to have lost its power of coagulation.

Position of ticks on the host. In the immense majority of cases the ticks are situated along the line of the spinal column, both in the natural and in the experimental cases.

The following are given as probable reasons why the ticks select the line of the back as their point of attachment:—

1. There is frequently a parting in the wool along the back.
2. The ticks are likely to be brushed off on to the back when the animals pass under bushes.
3. All the denuded portions of a sheep's body are greasy.
4. The wool is very thick on all other parts of the body except on the brisket where ticks are occasionally found.

In animals other than sheep, including man, the favourite seat of attachment appears to be the neck, but in horses they are found in the perineal region also.

Of 87 ticks collected from sheep 53 were females and 34 males.

In view of the fact that the male tick frequently attaches itself without any females being present, it is more dangerous than the male of any other species—if the male is capable of producing the disease, a point which requires elucidation.

(78) HADWEN (S.) & NUTTALL (G. H. F.). **Experimental "Tick Paralysis" in the Dog.**—*Parasitology*. 1913. Oct. Vol. 6. No. 3. pp. 298-301.

The dog used in this experiment had been used fourteen months previously for a piroplasmosis experiment, and had made a complete recovery after treatment with trypanblue.

The ticks used were collected in June 1913 and on July 8th a single female was placed upon the dog. On the following day the tick attached itself on the right side of the neck just in front of a line joining the scapular angles. On the ninth day symptoms of paralysis appeared, the hind quarters being affected. Sensation in the hind legs was unimpaired. On the following day the fore limbs were affected, and the dog could only raise its head a little. There was incontinence of urine and loss of appetite. During the next three days the breathing was laboured and the animal had to be fed, the paralysis persisting. From the 14th day the symptoms began to subside and by the 16th day the dog was as well as ever. On the 18th day the tick was removed about two-thirds engorged.

On July 25th a single female was placed each upon a horse and a jackal. Both ticks attached themselves, but did not engorge to repletion and failed to produce any effects.

In 1912 a number of experiments were carried out with guinea-pigs and some ticks obtained from eggs of *D. venustus*.

In the first experiment larvae and then nymphs were placed upon guinea-pigs, but the result was entirely negative.

In the second experiment larvae were placed on a guinea-pig and they dropped off engorged in from 3 to 6 days. Eight days later nymphs began to emerge. These were placed upon a guinea-pig which died on the 6th day after their application.

In the third experiment larvae were placed upon a guinea-pig which died on the 5th day. Some of the nymphs obtained were placed upon a guinea-pig which died on the 7th day. Others were placed upon a ram's scrotum without producing any effect. The adults were also without any effect when placed upon a ram's scrotum.

(79) WILLIAMS (A. J.). Notes on an Outbreak of Horse Sickness connected with the Presence of *Lyperosia* as a possible Transmitter. —*Veterinary Jl.* 1913. Aug. Vol. 69. No. 458. pp. 382-386

In this article the author describes the occurrence of an outbreak of horse sickness under conditions which suggested that *Lyperosia* might be the transmitting agent. The cases occurred at Dilling and later at El Obeid, in November and December 1908.

The first two cases occurred in animals that had been at Dilling for sometime, and the subsequent cases among the horses of a squadron of cavalry which had arrived there from Kordofan.

The first case occurred among the cavalry horses on the 6th day after the first horse died and the 7th after their arrival. The second case in this batch of animals occurred a week later.

The horses were then moved to El Obeid, ninety miles away. There seven cases occurred and it appeared to be probable that in five of these the infection was contracted before the horses left Dilling. After their departure for El Obeid three further cases occurred at Dilling, making 14 in all.

Attention is drawn to the following points:—

No cases occurred among horses purchased locally. The symptoms and course of the disease were absolutely typical. Horse sickness is known by the Arabs to occur in the district, which is sandy and desert-like, and where during the dry season water is obtained from wells 60 feet deep. The disease is known to make its appearance with the first rains and to disappear with the onset of the cold season. Enquiries shewed that several cases had occurred in the locality and it was considered to be unusual that cases should occur so late in the year.

There were no mosquitoes in the district, and the only biting fly found was *Lyperosia minuta*, and the author thinks that taking all the circumstances into consideration it appears to be not improbable that this fly was the transmitting agent, carrying the infection directly from horse to horse.

(80) KUHN (P.). Bemerkungen zu der Arbeit von Dr. H. Sieber "Experimentelle Untersuchungen über die Pferdesterbe." [Observations on Dr. Sieber's Article "Experiments in connection with Horse Sickness."]—*Zeitschr. f. Infektionskrankh. parasit. Krankh. u. Hyg. d. Haust.* 1913. Oct. Vol. 14. No. 4/5. pp. 316-320.

This paper contains some criticisms of an article published by SIEBER in the same journal in 1911 (Vol. 10, No. 2/3).

- (81) JOWETT (W.). **Pulmonary Mycosis in the Ostrich.**—*Jl. Comp. Path. & Therap.* 1913. Sept. Vol. 26. No. 3. pp. 253-257. With 1 text-fig.

Although the author was unable to carry out experiments to identify the causal organism encountered in the lesions described in this article, the fact that the lesions differed markedly from those encountered by ARCHIBALD in the Sudan (see this *Bulletin* Vol. 1, No. 5, 1913, p. 316) appeared to warrant the publication of a note.

The cases occurred in birds from three to five years old and the period of visible illness was probably very short, though no definite information was forthcoming on this point. The lesions were found in the lung tissue only, the trachea and bronchi being free from macroscopic alterations. The lungs were congested and hepatised, and on section were found to be crowded with small yellowish tubercles rather larger than pins' heads. The tubercles were shot-like in consistence and could not be enucleated from the surrounding lung tissue.

On section they were found to be composed of three zones—a central necrotic zone containing numbers of thick-walled, branching, segmented filaments, a middle zone composed of leucocytes and epithelioid cells, through which many mycelial filaments passed in a radiating manner, and a peripheral zone in which the lung tissue shewed evidence of croupous inflammation.

The filaments measured from 2 to 4 microns in diameter and the older ones, which had very thick walls, resisted the penetration of stains. Free spores were fairly numerous and measured up to 4 microns in diameter.

"Mouldy" grape husks were thought to have been the vehicle of infection.

- (82) VINCENZO (Z.). **Sul *Criptococcus farciminosus* di Rivolta.** [The *Cryptococcus* of Rivolta.]—*Il Moderno Zooiatro.* Parte Scientifica. 1913. Sept. 30. Vol. 24. No. 9. pp. 381-385.

This paper contains a general survey of the author's observations regarding the nature of the organism, and he concludes that the parasite is not a protozoon but that it belongs to the saccharomycetes.

- (83) MARSHALL (C. H.). **Experiments with the *Micrococcus melitensis*.**—*Jl. London School Trop. Med.* 1913. Nov. Vol. 2. No. 3. pp. 220-223.

This short paper contains the records of a few experiments carried out with the object of increasing the virulence of the organism and to test the effect of "606" upon the course of the disease.

The author's conclusions are as follows:—

A few passages of the *Micrococcus melitensis* through the guinea-pig are insufficient to naturally increase the virulence for that animal.

The slightly increased virulence obtained by a few passages of the organism through guinea-pigs does not increase the virulence of the strain for rabbits.

The injection of "606" appears to have a definite inhibitory effect on *M. melitensis* when injected into the experimental animal shortly after inoculation with the organism.

When the original dose of "606" is insufficient to prevent infection a subsequent dose appears to have a definite curative effect.

REPORTS.

- (84) **NYASALAND PROTECTORATE. Annual Report on the Department of Agriculture for the Year ending March 31, 1913. Veterinary Division.** (GARDEN, E.)—Zomba: Printed at the Government Press. [Price 6d.]

This report deals with (1) The Veterinary Congress on Rinderpest held at Bulawayo (the resolutions passed are printed verbatim), (2) Trypanosomiasis, (3) Piroplasmosis, (4) Demodectic Mange, (5) Black Quarter, (6) Ophthalmia in Cattle, (7) Swine Fever, and (8) Dipping. The specimens examined during the year are given in a tabular statement.

Four outbreaks of trypanosomiasis are recorded, principally on or near the Zomba-Blantyre Road. No details regarding the magnitude of the outbreaks or the causal trypanosome are given.

Theileria parva is known to occur in the Protectorate and the presence of *B. mutans* is strongly suspected.

The spread of demodectic mange among [cattle has rendered notification necessary. Slaughtering of the affected and regular dipping of the whole herd is advocated as the only means of eradicating the disease.

Although the presence of Black quarter in the Protectorate is suspected, proof of its existence has not yet been furnished.

Two forms of ophthalmia in cattle are met with, viz., "Specific" which has been dealt with in a previous report, and "Verminous."

Verminous ophthalmia is caused by nematode worms which measure 10-14 mm. (female) and 20-24 mm. (male) in length.

"The parasites are readily seen gliding over the surface of the eye and in between the folds of the mucous membrane."

They are often found in tangled masses under the membrana nictitans.

Swine Fever made its appearance in the Protectorate in October 1912, the infection being traceable to Rhodesia. Fortunately the area involved was restricted, and owing to the villages being widely separated the disease died out.

Attention is emphatically drawn to the necessity of establishing tanks and undertaking systematic dipping for the eradication of ticks and tick-borne diseases.

- (85) **SOUTHERN RHODESIA. Report of the Chief Veterinary Surgeon (SINCLAIR, J. M.) for the Year 1912.**

African Coast Fever. During the year five outbreaks occurred, as compared with eight in 1911 and eighteen in 1910. The total mortality including animals slaughtered on account of a rise of temperature was 505. Of these 409 occurred at one centre.

Dipping has been found to be of immense value in controlling this and other diseases. The value of the method of dealing with the disease is evidenced by an account of the occurrence of the disease on the Umtali commonage. After a period of freedom lasting for several years a case occurred in January 1910. Three-day dipping was

resorted to and, although the cattle were not removed from the infected veldt, only four further deaths have occurred in two years.

Contagious Pleuro-pneumonia. A single case of this disease occurred at a farm close to the Bechuanaland Protectorate border, and since the last case of the disease recorded in the whole Territory occurred in 1904 it appears to be probable that it was introduced from Bechuanaland where it has existed for years. Quarantine and restriction of movement regulations were in force for three months and no further outbreaks occurred.

Anthrax. Two outbreaks occurred involving the loss of fourteen pigs, one donkey, and six head of cattle. The only record of a previous outbreak of anthrax in Southern Rhodesia is that which occurred at Ramaquabane in 1898.

Bovine Plasmoses. The immunity conferred by an attack of redwater gradually declines unless reinforced by constant re-infection by means of ticks. It has been observed that the dipping practised for the eradication of African Coast Fever has resulted in a decrease in the immunity against redwater and gall-sickness (anaplasmosis). It has also been observed that the strain of anaplasma has increased in virulence during the last year or two.

In order to establish immunity in imported stock the local virus must be used. The virus obtained from Pretoria did not confer complete immunity.

Black Quarter. No recurrence of the disease is reported.

Ovine anaplasmosis. The existence of this disease in various districts throughout the Territory has been demonstrated. The symptoms are anaemia, cachexia, and oedema of the intermaxillary space (bottle jaw).

Trypanosomiasis. Although tsetse fly exist to a considerable extent in the Sebungwe, Lomagundi, and Hartley districts, the diseases transmitted by them to domesticated animals are not of great economic importance from a veterinary point of view.

The only outbreak of any importance recorded during the year was the loss of 20 head of native cattle in the Sebungwe district. It is only in grossly infested areas that stock farming is impossible. Encouraging results have been obtained in the treatment of trypanosomiasis by the Veterinary Bacteriologist (BEVAN), but it is not thought advisable to publish an account of the method of treatment at present.

Importation of cattle from North-Eastern Rhodesia was suspended owing to the possibility that the animals might convey the human trypanosome, and it was suspended from North Western Rhodesia on account of the existence of "Shesheke cattle sickness" which appears to be a fatal form of trypanosomiasis occurring in an area free from tsetse flies.

Rabies. Outbreaks have occurred in eight or nine districts, but no figures are given. It is said that there is good reason for believing that the disease exists among the wild carnivora.

Horse Sickness. No statistics are available, but not more than a dozen deaths were reported.

(86) SOUTHERN RHODESIA. Report of Veterinary Bacteriologist (BEVAN, L1. E. W.) for the Year 1912.

Bovine Plasmoses. See Abstract 478, Vol. 1 of this *Bulletin*.

Horse Sickness. Experiments regarding immunisation against this disease have been carried out on fifteen animals and seven of these have withstood inoculation with virus.

Trypanosomiasis. See Abstract 487, Vol. 1 of this *Bulletin*.

In that abstract it is stated that further reference will be made to the trypanosomes appearing in two of the rabbits inoculated.

These rabbits were inoculated, one from a dog and one from a sheep, and a trypanosome of the *pecorum* type made its appearance in the blood. These trypanosomes were carried on in sub-inoculations.

A number of cattle were detained at Feira on account of the possibility of their conveying human trypanosomiasis. Three dogs and nine sheep were inoculated from them. Two of the dogs died of an infection of the *pecorum* type in 28 and 44 days. Four sheep became infected with a mild disease caused by a trypanosome of the same type. "In none of the animals have trypanosomes of the nagana type been encountered. In view of the large percentage of buck found by Kinghorn and Yorke to be infected with the so-called *T. rhodesiense* in the Luangwa Valley, this result is somewhat significant."

- (87) UGANDA PROTECTORATE. **Annual Report of the Department of Agriculture for the Year ended March 31, 1913.** Report of the Chief Veterinary Officer (HUTCHINS, E.), 1913. Kampala: Printed by the Uganda Company Press.

East Coast Fever. Owing to pressure of other duties little has been done during the year regarding this disease save mapping out areas in which it is endemic.

Rinderpest. During the year rather over 38,000 cattle in the Eastern Province were immunised against this disease. Approximately 30,000 of these were treated by the mixed method, a dose of virulent blood being administered at the same time as protective serum. The mortality seldom exceeded 5 per cent.

Trypanosomiasis. *T. pecorum* is responsible for the heaviest losses. In certain districts half the affected herds were carried off by the disease and there appears to be good reason for believing that in Bugerere and Buruli the disease is of recent introduction. The Eastern Province is particularly free from trypanosome infection but so far as is known there is nothing in the distribution of the biting flies which would account for this, but possibly the absence of big game is the real explanation.

Epizootic lymphangitis is widely prevalent and it is said that it can be arrested if treatment is undertaken in time, but it is doubtful if a permanent cure is ever effected.

Two fatal cases of horse sickness were seen during the year.

Restrictions have been imposed upon imported dogs, as rabies has been reported as occurring in jackals in British East Africa.

Trypanblue has proved to be effectual in the treatment of the majority of cases of piroplasmosis in the dog.

- (88) UGANDA PROTECTORATE. **Report of the Tour in South Africa, undertaken by RICHARDSON (U.F.), Veterinary Officer.** [MS.]

The tour described in this report was undertaken for the purpose of studying (1) East Coast Fever, particularly methods of inocula-

tion. (2) Trypanosomiasis. (3) Diseases of stock occurring in South Africa which may become of economic importance in Uganda. (4) South African methods of regulating cattle transport.

East Coast Fever. The importance of this disease in Uganda lies in the facts that: (1) There are parts of the country which have never suffered from the disease and others in which it is endemic, and cattle removed from the one to the other die from it. (2) In some parts of the country the raising of young stock is severely handicapped by East Coast Fever occurring in young calves (Amakebbe) and causing a mortality of from 1 to 80 per cent., according to the district. (3) Imported cattle quickly die of the disease. Reference is made to the procedures adopted in Southern Rhodesia and the Union of South Africa for dealing with the disease. In the former quarantine, movements of cattle to clean pastures and dipping are said to have given fairly satisfactory results. The same plan associated with slaughter of affected animals is giving good results in the Union. It is suggested that where dipping is found unsatisfactory it is probably due to want of care. Inoculation with spleen and gland pulp is said to give very irregular results, the reason for which is unknown.

Pulp made from the organs of an animal actually dying from the disease is believed by the officers in the field to be too powerful a material to be used as a vaccine. It is believed that the best material is obtained from an animal just after the remission of temperature. Richardson believes that inoculation might be resorted to with advantage in Uganda, but that it does not appear to offer much hope of dealing with Amakebbe.

Trypanosomiasis. With regard to the transmission of trypanosomiasis, it is said to be the general opinion of veterinary officers in Portuguese East Africa and Rhodesia that permanent fly areas are kept infective by tsetse flies, and that cattle which have become infected in these areas act as reservoirs outside the fly belts, the transmitting agents there being Tabanidae.

BOOK REVIEW.

- (89) HOLMES (Major J. D. E.). [M.A., D.Sc., M.R.C.V.S., Imperial Bacteriologist.] **A Description of the Imperial Bacteriological Laboratory, Muktesar; Its Work and Products.** 47 pp. With 33 plates. 1913. Superintendent Government Printing. Calcutta. India. [Price 8 annas, or 9 pence.]

This little volume is full of interest and instruction, particularly to those who have not had the opportunity of working in India. The difficulties encountered in the application in the field of the results of the laboratory investigations are clearly shewn, and the manner in which these difficulties have been overcome to a great extent plainly indicates the value of the laboratory work.

The book is divided into five chapters dealing with (1) A Short History of the Laboratory, (2) A Description of the Laboratory Site and Buildings, (3) The Research Work and Publications of the Laboratory, (4) The Practical Application of Serum and Vaccines, and (5) The Products of the Laboratory.

The chapter on the Research Work and publications will be found of great value when it is desired to refer to the literature concerning the diseases of stock in India.

Chapters 4 and 5 deal in a concise manner with the preparation and practical application of the various sera and vaccines prepared at the laboratory, and it is from these chapters particularly that a clear idea is obtained of the immense difficulties that have been and are being overcome.

The half-tone plates give an excellent idea of the general arrangement of the laboratory and its equipment, shewing especially the size to which the Station, which was opened in 1895 with a laboratory, post-mortem house, cattle shed, bungalow, and cattle-kraal, has grown.

RECENT LITERATURE.

[Continued from *Bulletin* Vol. 1, No. 5, pp. 321-326.]**Babesiasis (Piroplasmosis).**

- (90) MARTOGGIO (F.), STELLA (V.), & PROVENZALE (F.). Sulla Recettività dei Bovini somali verso il *Piroplasma bigeminum*.—*Ann. d'Igiene Sperimentale*, 1913. Vol. 23. (New Ser.). No. 3, pp. 315-324. With 3 charts.
- (91) NUTTALL (George H. F.). The Herter Lectures. III. Piroplasmosis.—*Parasitology*, 1913. Oct. Vol. 6. No. 3, pp. 302-320. With 14 text figs.
- (92) SABRAZÈS (J.) & BONDEAUD (Aug.). Note sur Quelques Cas Piroplasmose canine observés dans le Sud-Ouest de la France.—*Gaz. Hebdomadaire des Sci. Med. de Bordeaux*, 1913. Oct. 12. Vol. 34. No. 41, pp. 490-491.
- (93) THOMSON (J. G.) & FANTHAM (H. B.). The Culture of *Babesia (Piroplasma) canis* in Vitro.—*Ann. Trop. Med. and Parasit.* 1913. Dec. Vol. 7. No. 4, pp. 621-630. With 5 text figs. and 1 plate.

Epizootic lymphangitis.

- (94) HARBER (A. F.). Epizootic Lymphangitis and its Treatment.—*Veterinary J.*, 1913. Sept. Vol. 69. No. 459, pp. 408-414.
- (95) KENNEDY (W.). Epizootic Lymphangitis.—*Agric. J. of B. E. Africa*, 1913. March. Vol. 5. Part 1, pp. 43-47.

Filariasis.

- (96) STEFKO (W.). La Filariose des Oiseaux de la Russie Centrale.—*Bull. Soc. Path. Exot.*, 1913. Oct. Vol. 6. No. 8, pp. 592-594.

Leishmaniasis.

- (97) CANNATA (S.). Sul Reperto del Parassita di Leishman nel Sangue periferico. [The Detection of Leishmania in the Peripheral Blood].—*Malaria e Malat. d. Paesi Caldi*, 1913. Aug.-Sept. Vol. 4. No. 5, pp. 303-306.
- (98) CARONTA (G.). Sul Potere complementare del Siero di Sangue nella Leishmaniosi infantile. [The Complement Content of Blood Serum in Infantile Leishmaniasis].—*Malaria e Malat. d. Paesi Caldi*, 1913. Aug.-Sept. Vol. 4. No. 5, pp. 309-313.
- (99) SCORDO (F.). Alterazioni morfologiche dei Corpi del Leishman nel Kala-Azar. [Morphological Changes in Leishman's Bodies in Kala-Azar].—*Malaria e Malat. d. Paesi Caldi*, 1913. Aug.-Sept. Vol. 4. No. 5, pp. 313-317.
- (100) SPAGNOLIO (G.). Sul la Ganglio-puntura nella Diagnosi di Leishmaniosi. [Gland Puncture in the Diagnosis of Leishmaniasis].—*Malaria e Malat. d. Paesi Caldi*, 1913. Aug.-Sept. Vol. 4. No. 5, pp. 306-308.

Rabies.

- (101) MEJÍA (Carlos Ramos). Rabia y Salvarsan.—*Semana Medica*, 1913. Dec. 4. Vol. 20. No. 49 (No. 1038), pp. 1315-1318.
- (102) MOON (V. H.). The Organism of Rabies and Experiments in its Artificial Cultivation.—*Jl. Infectious Diseases*, 1913. Sept. Vol. 13. No. 2, pp. 232-235.
- (103) POOR (D. W.) & STEINHARDT (Edna). A Study of the Virus of Rabies, freed from the Cells of the Host and from Contaminating Organisms.—*Jl. Infectious Diseases*, 1913. Sept. Vol. 13. No. 2, pp. 203-231.

Rinderpest.

- (104) HOLMES (J. D. E.). Rinderpest. Further Investigations on Questions connected with the Economical Production of Antiserum.—*Memoirs Dept. Agriculture in India*. Vet. Ser., 1913. Nov. Vol. 2. No. 2, pp. 33–80.
- (105) OLIVER (E. W.). Note on Rinderpest.—*Bulletin* 28, *Agricultural Series*. Issued by the Dept. of Land Records and Agriculture, United Provinces of Agra and Oudh.
- (106) WÖLFEL (K.). Rinderpest in Deutsch Ostafrika. [Rinderpest in German East Africa.]—*Zeitschr. f. Infektionskrankh. parasit. Krankh. u. Hyg. d. Haust.*, 1913. Dec. 24. Vol. 14. No. 7, pp. 429–457. With 1 map.

Spirochaetosis.

- (107) KING (W. E.) & HOFFMANN (G. L.). *Spirochaeta suis*, its Significance as a Pathogenic Organism. Studies on Hog Cholera.—*Jl. Infectious Diseases*, 1913. Nov. Vol. 13. No. 3, pp. 461–498. With 1 plate and 3 text figs.
- (108) LEBAILLY (C.). Sur les Spirochètes de l'Intestin des Oiseaux.—*Compt. Rend. Soc. Biol.*, 1913. Nov. 21. Vol. 75. No. 32, pp. 389–391.
- (109) SWIFT (H. F.) & ELLIS (A. W. M.). A Study of the Spirochaetoidal Action of the Serum of Patients treated with Salvarsan.—*Jl. Experim. Med.*, 1913. Oct. Vol. 18. No. 4, pp. 435–449.

Trypanosomiasis.

- (110) HOLMES (J. D. E.). A Further Note on the Cure of Surra in Horses.—*Indian Civil Vet. Dept. Memoirs*. No. 3, pp. 73–77.
- (111) HOLMES (J. D. E.). Some Cases of Surra treated in the Field and in the Laboratory during the Autumn of 1911.—*Memoirs of the Dept. of Agriculture in India*. Veterinary Series, 1913. Aug. Vol. 2. No. 1, pp. 1–31.
- (112) KLEINE (F. K.) & FISCHER (W.). Schlafkrankheit und Tsetsefliegen. II. Mitteilung. [Sleeping Sickness and Tsetseflies.]—*Zeitschr. f. Hyg. u. Infektionskrankh.* 1913. Aug. 27. Vol. 75. No. 2, pp. 375–382.

Undulant Fever.

- (113) SÈJOURNANT (J.). La Fièvre Méditerranéenne en Algérie en 1912. [Undulant Fever (Malta Fever) in Algeria in 1912.]—*Ann. Inst. Pasteur*, 1913. Oct. Vol. 27. No. 10, pp. 828–838.

Biting Insects.**Flies.**

- (114) CARPENTER (G. D. H.). Second Report on the Bionomics of *Glossina fuscipes (palpalis)* of Uganda.—*Report Sleeping Sickness Commission Roy. Soc.*, 1913. No. 14, pp. 1–37. With 36 figs.
- (115) CHALMERS (A. J.) & KING (H. H.). The Distribution of *Glossina longipennis* (Corti, 1895).—*Jl. Trop. Med. and Hyg.*, 1913. Oct. 15. Vol. 16. No. 20, pp. 320–322. With 1 map.
- (116) FISKE (W. F.). The Bionomics of *Glossina*: a Review with Hypothetical Conclusions.—*Bull. Entomol. Research*, 1913. Sept. Vol. 4. Part 2, pp. 95–111.

Biting Insects—cont.

Mosquitoes.

- (117) THEOBALD (F. V.). New Culicidae from the Sudan.—[*Mucidus nigerrimus*, nov. sp., *Chrysconops nocturnus*, nov. sp., *Reedomyia sudanensis*, nov. sp., *Kingia maculoabdominalis*, nov. sp., *Aedimorphus quinquepunctata*, nov. sp., *Culicelsa centropunctata*, nov. sp., and *Heptaphlebomyia kingii*, nov. sp.]—*Ann. Trop. Med. and Parasit.*, 1913. Dec. Vol. 7. No. 4, pp. 591-602.

Ticks.

- (118) ROBINSON (L. E.) & DAVIDSON (J.). The Anatomy of *Argas persicus* (Oken, 1818). Part II.—*Parasitology*, 1913. Oct. Vol. 6. No. 3, pp. 217-256. With 4 plates and 8 text figs. (For Reference to Part I see Ref. 584, Vol. 1 this *Bulletin*.)

Protozoa.

- (119) O'FARRELL (W. R.). Hereditary Infection, with special reference to its Occurrence in *Hyalomma aegyptium* infected with *Crithidia hyalommae*.—*Ann. Trop. Med. and Parasit.*, 1913. Dec. Vol. 7. No. 4, pp. 545-562. With 3 plates.
- (120) RATZ (S. von). *Trichomonas* aus der Leber der Tauben. [Trichomonas in the Liver of the Pigeon.]—*Centralbl. f. Bakt.* 1. Abt. Orig., 1913. Oct. 4. Vol. 71. No. 23, pp. 184-189.
- (121) WENYON (C. M.). Observations on *Herpetomonas muscae domesticae* and some Allied Flagellates. With Special Reference to the Structure of their Nuclei.—*Arch. f. Protist.*, 1913. Sept. 25, pp. 1-36. With 3 plates, 6 text figs. and 1 diagram.

Unclassed.

- (122) COMINOTTI (L.). L'Anemia infettiva del Cavallo. [Equine Infectious Anaemia.]—*Clinica Veterinaria*, 1913. Nov. 30. Vol. 36. No. 22, pp. 983-996.
- (123) ELLIS (A. W. M.) & SWIFT (A. F.). The Effect of Intraspinal Injections of Salvarsan and Neosalvarsan in Monkeys.—*Jl. Experim. Med.*, 1913. Oct. Vol. 18. No. 4, pp. 428-434.
- (124) KNUTH. Weitere Beobachtungen über *Haemaphysalis punctata* als wahrscheinlichen Ueberträger des Erregers der inneren Verblutung (Milzruptur) beim Rinde. [Further Observations regarding *Haemaphysalis punctata* as the Probable Transmitting Agent of the Cause of Internal Haemorrhage (Rupture of the Spleen) in Cattle.]—*Berl. Tier. Wochenschr.*, 1913. Nov. 20 and 27. Vol. 29. Nos. 47, 48, pp. 832-833 and 853-854.
- (125) LEWIS (J. C.). Veterinary and Stock Report and some of the Diseases in Stock in the Northern Territory.—*Bull. of the Northern Territory*, 1913. July. No. 8, pp. 35. With 18 plates.
- (126) MITRA (Satyendra Nath). On a Peculiar Form of Pasteurella in an Indian Elephant.—*Centralbl. f. Bakt.* 1. Abt. Orig., 1914. Feb. 11. Vol. 73. No. 1, pp. 12-13. With 1 text fig.
- (127) MONTFALLET (D.). Contribution à l'Étude des Maladies du Sang du Bétail de Chile, 1913. Imprimerie Franco-Chilienne, Rue Villa Vicencio 315, Santiago de Chile. pp. 1-23.

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[No. 2.

BABESIASIS (PIROPLASMOSIS), NUTTALLIASIS, AND ANAPLASMOSIS.

- (128) CARPANO (M.). Kultur der Pferdepiroplasmen und Betrachtung über die Natur der Anaplasmen. [The Cultivation of the Piroplasms of the Horse and Views regarding the Nature of Anaplasms.]—*Centralbl. f. Bakt.* 1. Abt., Orig. 1914. Feb. 11. Vol. 73. No. 1. pp. 42-53. With 1 plate.

The following is the method employed for the cultivation of both *Nuttallia equi* and *Piroplasma caballi*.

Tubes are prepared containing 1 cc. of distilled water in which sodium chloride and sodium citrate have been dissolved, each in the proportion of 7 per cent. To these are added 9 cc. of fresh defibrinated blood containing the parasites. The tubes are incubated at 25° C.

The author has not been able to get undoubted multiplication of the parasite in the case of *Babesia caballi*. After 48 hours incubation numbers of the pear-shaped parasites are extracellular. Certain changes are to be observed in their chromatin. In some of the parasites the chromatin forms a small round compact mass, while in others it has a loose texture.

By the fifth to the eighth day regular and irregular star-shaped parasites may be seen. The rays of the stars are formed either of cytoplasm alone or partly of chromatin. At the same time there make their appearance large rounded elements, which are composed for the most part of faintly-stained cytoplasm. From the 10th to 15th day large irregular parasites possessing two pieces of chromatin can be found. One of the chromatin masses is large and usually round, the other is small and frequently thread-like.

Subsequently no further development takes place and after a variable length of time the parasites degenerate, the medium at the same time becoming completely haemolysed.

The author considers that these forms must represent sexual forms.

In cultures *Nuttallia equi* does not retain its vitality so long as *B. caballi*, but certain morphological changes and definite multiplication can be observed.

During the first 48 hours the large and medium-sized parasites which are present in the circulating blood almost entirely disappear, and cross-forms are very seldom seen. The bulk of the parasites

are small and are composed for the most part of chromatin, and indeed some appear to be composed of chromatin entirely.

The process of multiplication is one of direct division in which the chromatin first divides and subsequently the cytoplasm. Corpuscles containing 1, 2, 3, or 4 parasites may be seen. In the latter case they are very small and their arrangement recalls the classical cross-forms. Free forms containing one or two chromatin masses are not infrequently found.

By the 3rd or 4th day the parasites appear to have become even smaller, and shew a tendency to arrange themselves at the periphery of the red corpuscles.

About the 6th to 8th day very small rings and bacillary forms are to be found, and about the same time forms occur possessing the morphological features of anaplasms, but in some of these a rim of cytoplasm can be detected. Parasites which have undergone division either escape from the red cells or are set free from them by their disruption, and would appear from stained preparations to enter fresh ones.

Free forms of the parasite are motile.

By the 20th to 30th day practically all the parasites present the morphological characters of anaplasms, and by this time haemolysis is almost complete.

The author had only one doubtful success in his attempts to transfer the cultures to fresh tubes.

The author has found the anaplast-like parasites in horses suffering from and recovering from nuttalliasis, and he has also seen cases in which only the anaplast forms were to be found.

In the portion of the paper dealing with the nature of anaplasms the author summarises at some length the views expressed by various authors, and concludes that anaplasms cannot be considered as absolutely unconnected with the life-cycles of piroplasms.

(129) CARPANO (M.). *Piroplasmosis equina*.—*Centralbl. f. Bakt.* 1. Abt., Orig. 1914. Feb. 11. Vol. 73. No. 1. pp. 13-40. With 3 plates and 6 text-curves.

In this paper the author describes in some detail his observations regarding the occurrence of two types of parasite in equine piroplasmosis in Italy. His description of the morphology of the organisms and their methods of multiplication closely agrees with that given by NUTTALL and STRICKLAND in 1912.

The author believes that *R. bursa* and *B. annulatus* are the transmitting ticks under natural circumstances, but has apparently been unable to furnish experimental evidence of this.

The clinical, pathological, and epizootiological characters of the two conditions are dealt with at some length.

The author's conclusions are as follows :

1. Equine piroplasmosis may be caused by two distinct parasites, one of which is of comparatively small size. This parasite is characterised by a special method of multiplication which results in the formation of four parasites arranged in the form of a cross. The name *Nuttallia equi* has been given to this parasite. The other parasite is of larger size, and occurs in the form of twin pear-shaped bodies. This parasite multiplies

in the circulating blood by a process of true budding. This organism is known as *Babesia caballi*.

2. The two parasites have a wide distribution, especially in the Old World. *Nuttallia* appears to have a wider distribution than *Babesia caballi*.

Both forms occur in Italy, and, of the two, *Nuttallia* has a wider distribution. Only *Babesia caballi* has been observed up to the present in Eritrea.

3. In Italy a large proportion of the native horses possess a certain degree of immunity, which appears to be partly natural and partly acquired. Imported horses are very susceptible.

4. There are no very marked distinguishing features between the clinical and pathological features presented by the diseases caused by the two types of parasite. There are certain characters of an epizootiological nature which are special to the two diseases.

5. Under natural conditions the diseases are transmitted by *Rhipicephalus bursa* and *Boophilus annulatus*, the latter being responsible for the transmission of *Babesia caballi*.

The diseases are transmissible by blood inoculation, and in cases where failure results this is due to immunity of the animals used.

6. When an animal has recovered from one of the forms of the disease it remains immune to it for a variable length of time. Owing to this fact it has been possible to separate the two types of parasite. An animal that is immune to *Nuttallia equi* can be infected with *Babesia caballi* and vice versa.

- (130) SCHELLHASE (W.). Ein Beitrag zur Kenntnis der Piroplasmosis der Schafe und Esel. Therapeutische Versuche mit Trypanblau. Ueber die Anaplasmosis der Esel. [Piroplasmosis of the Sheep and Donkey. Therapeutic Experiments with Trypanblue. Anaplasmosis of the Donkey.]—*Zeitschr. f. Infektionskrankh. parasit. Krankh. u. Hyg. d. Haust.* 1914. Feb. Vol. 15. No. 1. pp. 93-97.

The experiments recorded in this paper were carried out with the object of investigating the course of the disease in cases of acute piroplasmosis, the susceptibility of the goat to the piroplasm of the sheep, and the effect produced by trypanblue, if any, upon the course of the disease in the acute stages.

Two young and clinically healthy goats and sheep were inoculated subcutaneously with 20 cc. of defibrinated blood from a sheep infected with piroplasmosis. Parasites were comparatively numerous in the blood.

A few anaplasms were found in the blood of one of the goats before inoculation but no piroplasms. The details of the experiments are a little difficult to follow as the results of daily examinations of the blood are not given.

Apparently anaplasms were first observed in the blood of goat 1 on the 8th day, and piroplasms three days later.

The blood of goat 2 which contained anaplasms at the time of inoculation, was found to be negative on the following day and on the 6th day after inoculation. On the 9th day anaplasms again appeared, to disappear the following day.

Sheep 1 had piroplasms in its blood on the 1st, 9th, 10th and 14th days after inoculation.

Sheep 2. Piroplasms were found on the 8th day and again on the 14th day.

The temperatures of the animals ranged from 38.5° to 41.3° C.

The author concludes from these experiments that piroplasms and anaplasms may be present in the blood of clinically healthy sheep, and suggests the probability that the majority of sheep and goats contract piroplasmosis and anaplasmosis during early life, and either recover or die. It is due to this fact that the experiments had doubtful results.

Piroplasmosis of the donkey.—Up to the present occasional cases only of piroplasmosis of the donkey have been discovered in German East Africa. The author has recently encountered two actual outbreaks of the disease. In one instance 4 out of 11 animals were found to be affected, and in another 10 out of 14. About 25 out of 75 donkeys died during the rains (principally foals) and five mares aborted. In the donkey the disease generally takes a chronic course. The piroplasms are rather longer than *Piroplasma mutans*, and the chromatin is relatively large in amount; "ring," "note," "pear-shaped," and "match-like forms" occur. Parasites are generally scanty in the blood.

Effects of trypanblue.—Five emaciated donkeys were given 40 to 100 cc. of 2 per cent. trypanblue intravenously, but no curative effects were produced.

Anaplasmosis of the donkey.—In the blood of two of the animals treated with trypanblue bodies resembling anaplasms were found, in addition to the piroplasms. On a number of occasions the appearance presented by the bodies suggested multiplication.

TRYPANOSOMIASIS.

(131) LEESE (A. S.). Preliminary Report on Military Camels in Jubaland.—Manuscript Report forwarded from the Colonial Office on April 23rd, 1914.

The author was detailed to proceed to Kismayu, Gobwen, and Serenli, to investigate the causes of mortality and unthriftiness in camels.

At Gobwen, where the author spent six days, there were 30 riding-camels of the Camel Corps, and of these at least 30 per cent. were affected with trypanosomiasis, clinically indistinguishable from surra. Advice was given regarding the suitability of certain grazing grounds and the diet of the animals. At Serenli, where there were 80 camels, only 10 were found to be affected with trypanosomiasis. Owing to continual movement it was impossible to trace the source of infection, but three of the infected animals were remounts only just arrived from Gobwen.

The principal causes of the mortality, amounting to 35 per cent. per annum, were the unfavourable conditions to which the animals were subjected on arrival in Jubaland, resulting in loss of condition, exhaustion, and death due to pneumonia, lung-abscess, and enteritis.

The unfavourable conditions were :

1. Putting animals to hard work when not in fit condition.
2. Working immature animals.
3. Particularly active patrol work, rendering it impossible to study the needs of the camels in so full a manner as under ordinary conditions.

4. Apathy on the part of the men regarding their animals.
5. Working camels hard through the hot season.
6. The feeding on an insufficient quantity of uncrushed grain, badly infested with weevils.
7. Absence of veterinary advice and suitable food for sick animals.
8. The frequency of sore backs.

Most of the animals were in very poor condition and there were many cases of necrosis of the skin, a disease termed "Gub" (or in India "Joolak"). In most cases the poor condition was not due to disease.

The cases of trypanosomiasis were isolated; one which had also a lung abscess was destroyed. Nine of the animals were rested for four months and then used for safari. Eleven of the total number of animals were $5\frac{1}{2}$ years old or under, and these were kept from hard work until the second pair of permanent incisors were cut (rising 6 years old).

Fly surveys were made from Selagli to Lalashid and Gaba-hari (the northernmost outposts), over routes most frequently used, and warnings against certain camps and grazing grounds given.

Recommendations regarding the management of riding-camels.—

Arab camels require about 5 months to become acclimatised, and should be bought at about 5 years old, so that they may become acclimatised to the country and feeding in the East African Protectorate and be ready for work at rising 6 years old.

The corn ration should be 8 pounds when the animals are at work or grazing between safaris. Millet and grain are the best, particularly the former, as it can be obtained locally. If sim-sim cake, cottonseed, or maize are used, these should not form more than $\frac{3}{8}$ of the ration. All grain should be crushed. If given uncrushed, millet and grain should be first soaked. In the absence of salt-bush, $\frac{3}{4}$ ounce of crushed rock-salt should be given when not on safari. The salt should be given every third or fourth day (i.e. $2\frac{1}{4}$ to 3 ounces), on watering day, first thing in the morning.

No marching should be done between 10.30 and 3 o'clock in the cool season, and these hours should be earlier and later respectively in the hot season. The average daily distance for fit riding-camels on safari is about 20 miles. A good rest should be given before the next safari.

Camels should be trained to go for four days without water in the dry season.

Saddles should be so constructed as to prevent pressure on the spine. This pressure is the most frequent cause of sore backs. Among 200 transport camels at Serenli 64 cases of trypanosomiasis had been discovered up to date, and this is the principal cause of death. Grain is not necessary for transport camels if they are properly cared for and are not unduly pushed. The distance covered daily on safari should not exceed 15 miles, and a rest should be given before the next safari is undertaken. The marches should be averaged, provided water supplies permit.

For working Somali camels not getting grain, watering in the dry season should be every three days.

Well-grown camels can carry 300 pounds if well cared for and under proper conditions on safari.

Marching by rivers should be avoided whenever possible, especially during rains and for four or five weeks after. When it is necessary, the less valuable camels should be used, and marching should be carried out at night, the animals grazing in the desert.

It is probably only below Selaghi that travelling by river route is dangerous at all times.

A report on trypanosomiasis, the distribution of biting flies, and on other infective diseases and preventive measures will follow later.

- (132) YAKIMOFF (W. L.) & SCHOKHOR (N. I.). *Recherches sur les Maladies tropicales Humaines et Animales au Turkestan. (3) Les Trypanosomiasés des Chameaux et des Anes au Turkestan.* [Investigations regarding Human and Animal Tropical Diseases in Turkestan. (3) The Trypanosomiasés of the Camel and the Ass in Turkestan.]—*Bull. Soc. Path. Exot.* 1914. Mar. Vol. 7. No. 3. pp. 187-188.

The authors have examined 476 camels, and have found 22 infected, but they state that this figure is probably too low, as camels may be infected without the parasite being discoverable in the peripheral blood.

The parasite was in all cases transmissible to guinea-pigs, and is said to be of the surra-nagana type. The possibility is suggested that it is in reality the trypanosome of Indian surra. They have never observed trypanosomes in the peripheral blood of mules and donkeys in Turkestan, but they state that at Bukhara they took from 1 to 3 drops of blood from 216 donkeys, diluted it with physiological serum and inoculated a guinea-pig subcutaneously. The animal became infected, the trypanosome being of the same type as that found in the camels.

- (133) LAVERAN (A.). *L'Agent du Debab d'Algérie est le Trypanosoma soudanense (Laveran).* [The Causal Agent of Debab is *Trypanosoma soudanense*.]—*Compt. Rend. Acad. Sci.* 1914. Jan. 12. Vol. 158. p. 93.

The author has carried out a fresh series of experiments shewing that Debab is caused by *T. soudanense*, a trypanosome originally obtained from the Upper Niger.

Cross immunity experiments have been carried out. The strains used were the original *soudanense* strain sent to France in 1906 by CAZALBOU and since kept running in guinea-pigs, and the debab virus obtained from a dromedary from the Touggourt-Biskra district.

The animals employed were: A goat having a high degree of immunity to *T. soudanense*, a control goat, and a goat immune to surra, which according to certain observers is allied to debab.

The experiments had the following results:—

The goat possessing immunity for *T. soudanense* resisted infection with the trypanosome that is responsible for debab, while the control goat and that immune to surra became infected. In a previous publication the author shewed that cattle immune to debab resisted infection with *T. soudanense*, and in the present note evidence is furnished that a goat immune to *T. soudanense* resists infection with

debab. The demonstration of the identity of the viruses is therefore complete.

The strain of *T. soudanense* in the possession of the author is slightly more active than the Algerian strain of the debab virus, and this explains why some animals immune to the latter became infected by the former.

In order to avoid this source of error in carrying out cross immunity experiments for the identification of two viruses, the immunity towards each must be tested by the other.

(134) BLACKLOCK (B.) & YORKE (W.). The Probable Identity of *Trypanosoma congolense* (Broden) and *T. nanum* (Laveran).—*Ann. Trop. Med. & Parasit.* 1913. Dec. 30. Vol. 7. No. 4. pp. 603-607.

In a previous paper (see this *Bulletin* Vol. 1, p. 76) a description was given of two parasites found in the blood of a horse imported from the Gambia. These parasites were separated accidentally. One was certainly *T. vivax*, but regarding the identity of the other there was some doubt.

The authors succeeded in infecting small animals with this trypanosome and finally decided that it was *T. dimorphon* (*T. congolense*). They have been able to maintain this parasite in rats. The subinoculations made from the time of isolation up to the 57th generation are shewn in a table, and from this the following information may be gathered.

1. Most of the early inoculations failed, while subsequently they were invariably successful.

2. The course of the disease in the earlier generations was chronic, and in the later ones acute. The average length of life of the rats from the 5th generation (when the strain was firmly established in the rat) to the 14th, was 88.6 days, but the average duration of life of the rats in the 42nd to 51st generations was only 8.6 days.

This appears to the authors to have some significance in connection with the nomenclature of the parasite. *T. congolense* and *T. nanum* are morphologically identical. They are spread by the same species of fly, and flies are infected in the same manner by each parasite. Had only a small number of experimental animals been available, as is usually the case in the tropics, the trypanosome would probably have been identified as *T. nanum*. WEISSENBORN described a short aflagellar trypanosome of equines in Togoland in 1911 and stated that it was of inconstant virulence.

The Sleeping Sickness Commission of the Royal Society state that *T. pecorum* loses its pathogenicity for other animals by passage through goats, thus giving rise to the erroneous idea that a separate species, *T. nanum*, exists.

The Belgian Sleeping Sickness Commission have found that aflagellar trypanosomes occurring in dogs are not always pathogenic for the rat and guinea-pig.

The authors can therefore find no evidence warranting a distinction being made between the morphologically identical parasites *T. congolense* and *T. nanum*, on the grounds of pathogenicity.

- (135) TRAUTMANN (R.). *Inoculation Positive de Trypanosoma cazal-boui á un Cercopithecus patas*. [Successful Inoculation of a *Cercopithecus patas* with *Trypanosoma cazal-boui*.]—*Bull. Soc. Path. Exot.* 1914. Feb. Vol. 7. No. 2. pp. 118-121.

With the exception of a few cases of transient infection in rats and guinea-pigs, *T. cazal-boui* is as a rule transmissible to ruminants and equidae only. A *Cercopithecus patas* was inoculated subcutaneously with 5 cc. of blood containing large numbers of trypanosomes from a calf which was on the point of death.

On the third day there was distinct autoagglutination of the red corpuscles. Trypanosomes appeared in the blood in very small numbers on the 6th day. On the 9th day, when no trypanosomes could be found in the blood, a sheep, a native dog, a young guinea-pig, a cynocephalus monkey, two grey rats, and a rabbit were inoculated from it. Trypanosomes were not seen again until the 11th day. On this day the monkey died.

At the post-mortem, all the glands, the liver, spleen, and kidneys were found to be enlarged.

The blood shewed a few degenerating trypanosomes, but none were found in one of the mesenteric glands.

The dog, guinea-pig, cynocephalus monkey, and rabbit failed to become infected. The rats never shewed trypanosomes in their blood, but they died on the 12th and 18th days. The sheep became infected and trypanosomes were found on the 8th day. Two patas, a rabbit, and a guinea-pig inoculated from the sheep failed to become infected, but a kid died on the 20th day. The trypanosome was identified by MESNIL.

- (136) DELANOË (P.). *Des Variations du Pouvoir Infectieux et de la Virulence du Trypanosoma dimorphon, à partir d'Infections Naturelles présentées par les Boeufs et les Moutons. Note préliminaire*. [Variations in the Pathogenicity and Virulence of *T. dimorphon* derived from Natural Cases in the Ox and Sheep.]—*Bull. Soc. Path. Exot.* 1914. Jan. Vol. 7. No. 1. pp. 58-63.

During a period of rather less than 6 months the author has examined some 3000 cattle and sheep for trypanosomiasis. The examinations were made in the course of the inspection of animals passing down from Upper Senegal and the Niger to the coast. For the detection of trypanosomes a single examination of moist blood was made, the animals which were declared free from infection upon the result of this examination being allowed to proceed. The only trypanosomes encountered were *T. cazal-boui*, *T. dimorphon*, and *T. pecaui*. The latter was found in a few instances only.

A series of 7 sets of experiments were carried out with *T. dimorphon*. In five instances the trypanosome (derived from 3 oxen and 2 sheep) was not inoculable directly to either the rat or the guinea-pig. In one case the trypanosome was transmitted directly to the rat, but not to the guinea-pig, and in the remaining case it was inoculable to both the rat and the guinea-pig.

Details are given of the experiments, and from these it appears that rats and guinea-pigs may possess complete immunity against

T. dimorphon, repeated intraperitoneal inoculations with small quantities of virulent blood, and single intraperitoneal or intravenous inoculations with large quantities failing to break the immunity down.

In the instance in which the rat became infected while the guinea-pig proved refractory, the infection in the rat was a very slight one. Trypanosomes were found in its blood in very small numbers on the 21st and 81st days only.

The last series of experiments shewed that the trypanosome is capable of setting up a rapidly fatal infection in both the rat and the guinea-pig. In this instance two white rats and one out of the two guinea-pigs inoculated from the infected ox shewed a mixed infection of *T. dimorphon* and *T. pecaui*, and died. A goat, whose blood had failed to cause any infection when inoculated into a rat, was inoculated from the ox, and contracted the dimorphon infection only. From this goat five field rats and two guinea-pigs were inoculated with fatal results.

The author does not think that there are any specific differences between the trypanosomes without free flagellum which are directly inoculable to rodents and those that are not. There is no essential morphological difference.

In a future paper the author proposes to shew that he has been able to inoculate rats with the virus against which rats possessed a complete immunity at the first inoculation.

The author believes that there are several strains of *T. dimorphon* which differ from each other only in the varying susceptibility possessed by animals towards them.

- (137) LAVERAN (A.) & MARULLAZ (M.). *Essais d'Immunité contre le Nagana Expérimental des Souris*. [Attempts to immunise Mice against Experimental Nagana.]—*Bull. Soc. Path. Exot.* 1914. Jan. Vol. 7. No. 1. pp. 53-58.

The experiments of RONDONI and GORETTI have been repeated using trypanosomes subjected to salvarsan in very dilute solutions as the immunising agent. The strain used was the "ferox" strain (EHRlich).

In all, eight experiments were made, and six mice were used in each. The full details of four of the experiments are given and the remainder are summarised. The trypanosomes were obtained by centrifuging blood containing large numbers after the addition of citrated broth, and recentrifuging. The trypanosomes so obtained were mixed with the salvarsan solutions varying in strength from 1 in 1,000 to 1 in 40,000. These were left for half an hour at room temperature, centrifuged, washed with salt solution, and injected.

The results obtained differed in a marked degree from those of RONDONI and GORETTI. Whereas the latter authors obtained an absolute immunity in 40 to 50 per cent. of animals, Laveran and Marullaz have not obtained a permanent immunity in any instance.

In 30 instances a more or less rapid infection followed the immunisation. Of 18 cases in which vaccination did not cause infection, 16 became infected when tested, and two which resisted a first infection became infected when a second dose of active virus was administered.

A slowing of the movements of the trypanosomes in the salvarsan solutions is not a sufficient guide as to the effects of the salvarsan. Dilutions of 1 in 10,000 to 1 in 5,000 simply increased the period of incubation, and dilutions of 1 in 1,000 and 1 in 2,000 yielded the same results as dead trypanosomes, i.e. failed to produce either infection or immunity.

- (138) JOHNS (F. M.). *Trypanosoma americanum*.—*New Orleans Med. & Surg. Jl.* 1914. Jan. Vol. 66. No. 7. pp. 533-534.

While working upon the cultivation of *B. bigeminum* the author encountered a very large trypanosome in the blood of a calf, and subsequently found 43 adult animals and 2 yearlings to be infected, by direct examination.

It was found possible to cultivate the trypanosome by adding half the volume of 1 per cent. dextrose to defibrinated cow's blood and incubating at any temperature from 27° to 40° C. It was found that the addition of serum containing complement to the cultures after the first 24 hours destroyed them. The author concludes from this that if an insect transmitter feeds again before the trypanosomes contained in the infective feed reach the hind gut, the insect becomes non-infective.

- (139) DELANOË (P.). **Le Fonctionnement du Service de Prophylaxie à Bouaké (Côte d'Ivoire) à l'Égard des Trypanosomiasés Animales du 10 Juin au 31 Déc. 1913.** [The Working of the Prophylactic Measures against Animal Trypanosomiasis at Bouaké (Ivory Coast) from 10th June to 31st December, 1913.]—*Bull. Soc. Path. Exot.* 1914. Feb. Vol. 7. No. 2. pp. 152-160. With 1 map.

The new sanitary measures involve the systematic examination of the blood of animals passing through Bouaké. The rule adopted is, that only those animals in which trypanosomes cannot be found at a single examination are allowed to go to the coast. Affected animals are kept under observation at Bouaké, or if sufficiently fat are slaughtered.

Only three or four horses have been examined, the remaining animals being cattle and sheep coming for the most part from Upper Senegal and the Niger. Out of 4,086 animals (2,115 cattle, 1,971 sheep) only 214 came from the Ivory Coast. Bouaké is the centre of the cattle traffic, the herds from the Soudan being brought there by middlemen for transmission to the coast and other parts.

The animals are inspected before they are sold and are branded in a manner indicating whether they have passed the examination or not, so that the loss rests principally on those bringing the animals to Bouaké. Infected animals that are in sufficiently good condition to be slaughtered and consumed locally fetch considerably lower prices than those that are passed.

Animals may be rejected on account of emaciation alone, and to save time the blood of these is not subjected to examination.

Emaciated animals may have no trypanosomes in their blood and in

the blood of fat animals they may be numerous. This is explained by the fact that the number of trypanosomes present in the blood at different stages of infection varies. The meat from well-nourished infected animals does not differ in any way from that of healthy animals.

Twenty-one per cent. of the cattle were found to be infected, and 4.6 per cent. were condemned for emaciation.

Of the sheep 10.4 per cent. were condemned for trypanosomiasis and 0.8 per cent. for emaciation.

A table is given shewing the number of cattle and sheep examined each month during the period under review. The increase in the number of animals inspected during December as compared with the previous months indicates that the inspection has not hindered trade.

Of the three types of Soudan cattle (the Zebu or large humped animals, the small animals without humps, and the cattle with small humps), the first are most frequently found to be infected and they do not bear long journeys well.

The animals that are infected to the smallest extent, and are less frequently emaciated, are those from Lobi. The herds having the highest percentage of diseased and emaciated animals were those from Macina, where souma occurs in a very virulent form.

The only trypanosomes encountered were the *T. cazalboui*, *T. dimorphon* and the *T. pecaui*, in order of their frequency.

The author states that confirmation can be furnished of the part played by *Stomoxys* in the transmission of souma. The inspection has been criticised as hindering trade, but the author states that apart from any consideration of prophylaxis the inspection has lead to an improvement in the quality of the animals.

- (140) MESNIL (F.) & BLANCHARD (M.). Sur l'Identification du Virus d'un Cas de Trypanosomiase Humaine contractée au Laboratoire. [The Identification of the Virus in a Case of Human Trypanosomiasis contracted in the Laboratory.]—*Bull. Soc. Path. Exot.* 1914. Mar. Vol. 7. No. 3. pp. 196-200.

The case referred to in this paper is that of Professor LANFRANCHI of Parma.

It is said that only two strains of trypanosomes, namely, nagana and surra, had ever been kept at the University of Parma.

A strain of the trypanosome which was thought to be the cause of LANFRANCHI'S infection was obtained from Parma, and animals were inoculated from LANFRANCHI himself.

A number of investigations were carried out with these trypanosomes and the sera from infected animals, and the general results indicated that the animal strain received from Parma and that isolated from LANFRANCHI were quite distinct.

Cross immunity tests and the actions of protective sera indicated that the strain derived from Lanfranchi did not differ from *T. gambiense*, and that the animal strain derived from Parma was *T. evansi*.

The authors are not prepared to state definitely that the strain from the human source was actually *T. gambiense*.

- (141) ROUBAUD (E.) & LAFONT (A.). *Expériences de Transmission des Trypanosomes Humains d'Afrique par les Moustiques des Habitations (Stegomyia fasciata)*. [Experiments regarding the Transmission of the Human Trypanosomes in Africa by Mosquitoes (*Stegomyia fasciata*).]—*Bull. Soc. Path. Exot.* 1914. Jan. Vol. 7. No. 1. pp. 49-52.

The authors have used *T. gambiense* and *T. rhodesiense*, and monkeys, guinea-pigs, and rats were employed. It was found that transmission occurred in those cases only in which the mosquitoes were immediately transferred to susceptible animals from infected animals which had numerous trypanosomes in their blood. The authors point out, however, that the experiments are not strictly comparable with infection of man because of the hair covering the animals' bodies, this rendering it difficult for the mosquitoes to bite. The experiments also shewed that the virus does not remain active in the mosquito for more than 24 hours.

- (142) PRENTICE (G.). *Sleeping Sickness, Tsetse, and Big Game*.—*Brit. Med. J.* 1914. Feb. 7. pp. 293-294.

This paper contains a general summary of the author's experiences in connection with game and tsetse fly in Nyasaland and Rhodesia, where, save for three periods of furlough, he has worked since 1894.

He speaks with personal experience of large tracts of country where not a single fly was encountered years ago, and where now they abound. He attributes the spread of the fly to the increase in the number of game animals, and this in turn is attributed to the protective measures put into force by the Government. The lives of men and domesticated animals are being lost as a result of this spread of the fly.

The author advocates a general onslaught on wild animals as offering the best chance of eradicating trypanosomiasis. He does not believe that the destruction of the game would lead to the fly attacking man and domesticated animals, and expresses the opinion that with the destruction of the game the fly would disappear.

- (143) WECK. *Beobachtungen über Trypanosomen des Menschen und der Tiere am Rovuma-Flusse*. [Observations on the Trypanosomes of Man and Animals on the Rovuma River.]—*Arch. f. Schiffs- u. Trop.-Hyg.* 1914. Feb. Vol. 18. No. 4. pp. 113-124.

The question of the identity of the trypanosomes occurring along the Rovuma is of practical importance on account of the fact that *Glossina palpalis* does not occur along the course of the Rovuma from Mitomoni to Sasawara, but only *G. morsitans*. Although the latter fly has a wide distribution in German East Africa it appears up to the present to have been a source of danger to domesticated and wild animals only, no case of trypanosomiasis having been observed in man.

The trypanosome found along the Rovuma is said to be morphologically and biologically distinct from *T. gambiense*, and, according to TAUTE, identical with *T. rhodesiense*.

The view expressed by BRUCE that *T. rhodesiense* is identical with *T. brucei* is questioned, and it is said that while nagana is of common occurrence in wild and domesticated animals in the *Glossina morsitans*

district, cases of trypanosomiasis in man have not been observed. It should be noted that the diagnosis of nagana is based in the great majority of cases upon microscopic examination. The explanation of the absence of trypanosomiasis in man is possibly that *T. rhodesiense* requires certain climatical conditions, and, particularly, higher temperatures. There are however many areas in the colony with the same climatic and geographical condition where game is plentiful and *Glossina morsitans* occurs in large numbers, but in which trypanosomiasis has not been observed in man.

The possibility is not excluded that the trypanosome recognised as *T. brucei* in German East Africa is not the same as that occurring in Nyasaland and Rhodesia, and when diagnosis is based upon microscopic examination it is quite possible to confuse trypanosomes.

A number of species of wild and domestic animals have been found to be infected with trypanosomes by direct microscopic examination in the Mitomoni, Sasawara, and Lumesule Districts, and a number of positive experimental infections have been obtained.

Positive results were obtained in every case when monkeys were inoculated with blood containing the trypanosome occurring in man, no matter from what species of animal the infective material was obtained. In some cases an exaltation of virulence was observed when the trypanosome was carried on from one animal to another of the same species. Strains of trypanosomes which are not transmissible to the monkey are probably not identical with the human strain. The identity of the trypanosomes has been established by inoculation into various species of experimental animals, microscopic examination of living and stained specimens, and serum methods of diagnosis.

The author has found the human trypanosome to be inoculable for the monkey, dog, cat, rat, mouse, "huko" (a mole-like animal), guinea-pig, young leopard, and ngolombwe (a small antelope). With the exception of the last-named, all the infected animals died within a month. Only one strain of trypanosomes, found in a water-buck, was infective for the same experimental animals as the human strain. This was dimorphic, and intermediate forms were found. The long forms of the antelope strain were more active than the corresponding forms of the human strain, the movement appearing to be of one rapid vibration rather than a writhing one. In either case the movement involved little translation. In stained preparations marked displacement of the nucleus was observed in both strains. In the trypanosome found in the antelope the centrosome was frequently absent, but this was not observed in the human strain.

Cross immunity tests could not be carried out, but the activity of the serum was tested by LAVERAN and MESNIL's method in a number of cases, and certain biological differences were found between the two strains. It may therefore be concluded that the two are not absolutely identical.

The trypanosomes found in a ngolombwe failed to infect a monkey, but infected a rat. Inoculation of further rats from this one failed; the trypanosome therefore could not be *T. rhodesiense*. The antelope was inoculated with blood from an infected man containing numerous trypanosomes. The trypanosomes previously present in the blood did not increase in numbers. Two monkeys and a dog were then inoculated from the antelope and infection took place.

The author therefore concludes that the human trypanosome was present in the antelope's blood.

The author's conclusions may be summarised as follows :

The trypanosomes found in game in the neighbourhood of Sasawara resemble those found in man, but present certain morphological and biological differences which make it impossible to consider the two as identical.

The human parasite can be experimentally transmitted to game, and from this it may be concluded that the transmission occurs in nature through the medium of tsetse flies.

Killing off the game as a means of combating the disease must be avoided, as it would have the effect of driving animals having a slight infection into clean districts where the conditions are suitable for *T. rhodesiense*, and lead to the infection of flies, man, and animals. If the game is to be killed off, the extermination would have to be complete, a measure which is in the author's opinion impracticable.

Human beings must be considered as the most important carriers of infection. The disease was probably brought in from Portuguese territory by natives, and medical examination must therefore be established in order to detect the disease in natives entering the colony.

- (144) BECK (M.). Untersuchungen über ein am Rovuma (Deutschostafrika) vorkommendes Trypanosoma beim Menschen. [Investigations on a Trypanosome occurring in Man in the Rovuma District, German East Africa.]—*Arch.f. Schiffs- u. Trop.-Hyg.* 1914. Feb. Vol. 18. No. 3. pp. 97-101. With 1 plate.

In 1911 and 1912 a number of small foci of sleeping sickness were discovered along the Rovuma and its tributaries which must have been due to the introduction of the disease from the neighbouring Portuguese territory. Seventy-two cases have been discovered.

The disease appears to be transmitted solely by *G. morsitans*, and 8 to 10 per cent. of the flies were found to be infected with trypanosomes. It was impossible to identify the trypanosomes found in the flies owing to the lack of experimental animals.

Experiments were undertaken to ascertain whether the trypanosome occurring in man was identical with *T. brucei*, as cattle had died of *brucei* infection in the district.

The posterior extremity of the human parasite usually has a loose structure and stains faintly. The anterior end is pointed, the nucleus is round and oval, and it was never observed to lie posterior to the blepharoplast. In a few cases it was close to the blepharoplast, but this is due in the opinion of the author to uneven drying of the cytoplasm. The lack of a micrometer eyepiece rendered it impossible to make any comparative measurements.

A strain of *T. brucei* was obtained from an infected mule, and comparative experimental inoculations with the human and animal strains shewed that the human strain was rather more virulent for the dog, monkey and rat than the animal strain.

In the animals infected with the human strain long forms first appeared in the blood, these were followed by intermediate forms, and then by short thick forms. Irregular rounded forms were observed in the blood of rats inoculated with blood containing the human parasite.

For further differentiation of the parasites, agglutination and agglomeration experiments were carried out. The results given by the former failed to reveal any distinction between the two. Cross-

immunity experiments were also undertaken, and these were successful to the extent that the trypanosomes did not appear in the blood of immunised rats (rats infected and then treated with atoxyl) until from 10 to 15 days after they had appeared in the blood of rats that had not been treated.

A number of animals were examined for trypanosomes in infected districts with the following results :

Of 8 dogs examined 5 were found to be infected. Three goats were all negative. Two water-bucks were positive out of three examined, and one of two elands also contained trypanosomes. A gnu, leopard, a spotted hyena and a number of monkeys and birds were negative.

(145) DUKE (H. L.). **Wild Game as a Reservoir for Human Trypanosomes.**—*Brit. Med. J.* 1914. Feb. 7. pp. 289-292.

The author points out in the first place the great divergence of opinion held by observers engaged in the biological problems connected with sleeping sickness. The view held by the German investigators is that the game trypanosomes are not pathogenic for man, and that held by British investigators is that certain of the trypanosomes recovered from game are identical with *T. rhodesiense*.

Three years after the native population were moved from the islands and the lake shore of Victoria Nyanza, *G. palpalis* on the lake shore was capable of infecting monkeys with trypanosomes indistinguishable from *T. gambiense*, and an exactly similar trypanosome was recovered from the blood of two situtunga at about the same time or a little earlier. These animals were shot on Damba Island, a few miles from the mainland. The question arises whether the fly and the antelope trypanosomes are the descendants of the *T. gambiense* which was responsible for the epidemic, or are simply parasites of the antelope.

The author sets out his subject in the form of a number of questions and gives the available information upon which answers may be based.

With regard to the identity of the fly and the antelope trypanosomes, the author states that in a previous publication he has shewn that the identity cannot be doubted.

The evidence shews that the trypanosome of the fly is derived from the antelope and not from other sources.

Natives visiting prohibited areas are probably not the source of the infection of the flies and consequently of the antelope.

With regard to the question whether the lake-shore trypanosomes can be identified with *T. gambiense*, the answer is that while there is no good evidence to shew that the fly and the antelope trypanosome are not identical with *T. gambiense*, there is a certain amount of evidence to shew that they are.

Experiments shew that wild antelope of the species frequenting the lake shore are inoculable with the human trypanosome, and further that they are capable of acting as a source of infection for flies for 22 months at least, and probably longer.

The evidence on the subject of the infectivity of the wild fly to the wild-fly trypanosome is not so complete as could be desired, but it rather supports the view that the trypanosome of the wild fly is *T. gambiense* and not an antelope parasite.

There is no trypanosome known in Uganda possessing the animal reactions of the lake shore organism other than *T. gambiense*.

The percentage of antelope harbouring *T. vivax* and *T. uniforme*, that is, trypanosomes non-pathogenic for man, is slightly higher on the Island of Damba than on the mainland, 14.2 as compared with 10, but the natural conditions obtaining in two places are sufficient to account for this. The percentage of wild lake shore flies harbouring these parasites is about 0.1.

The author contends that the available evidence justifies the conclusion that the trypanosome recovered from the Damba situtunga and from the wild *G. palpalis* on the islands and on the coast of the mainland is a descendant of *T. gambiense*.

- (146) CIUCA (A.). Action des Abscès de Fixation sur la Trypanosomiase Expérimentale du Cobaye et sur son Traitement par l'Atoxyl. [The Effect of Fixation-Abscesses upon the Treatment of Experimental Trypanosomiasis in the Guinea-pig and its Treatment with Atoxyl.]—*Ann. Inst. Pasteur*. 1914. Jan. Vol. 28. No. 1. pp. 6-20.

The author refers briefly to the observations of a number of authors regarding the beneficial effects produced by abscess formation in protozoal diseases (malaria, canine babesiasis, and sleeping sickness).

The investigation was undertaken by the author because in the cases referred to the abscess formation was of an accidental nature.

Essence of terebene is one of the chemical substances which is specially used for the production of fixation abscesses. The majority of the experiments have been carried out with the Uganda strain of nagana, as, according to the author, the course of the infection in the guinea-pig with this trypanosome is very constant, trypanosomes persisting in the blood from their first appearance up to the time of death. A further reason for using this trypanosome is that, according to LAVERAN and THIROUX's experiments, atoxyl alone is unable to effect a cure. In some of the experiments the guinea-pigs were infected with surra.

Ordinary commercial essence of terebene was used and it was injected subcutaneously on the outer side of the thigh, as high up as possible in order to avoid gangrene of the foot, and diffusion of the pus during abscess formation. The injection caused extreme disturbance which lasted for about half an hour. The resulting inflammation reached its maximum about the 72nd hour, abscess formation occurring about the 4th or 5th day. The pus was thick and of a dirty white colour. A strong odour of terebene could be detected even when the abscess was left for 10 days before opening. The susceptibility of the guinea-pig to the terebene varied greatly. In some there was marked loss of condition in 10 days, in others no effect of this kind was observed, and in some guinea-pigs no local inflammation would be produced.

The dose varied from 0.5 to 1 cc.

A freshly prepared solution of atoxyl in sterile distilled water was used, and the dose varied from 1.5 to 2 cc.

The disappearance of the trypanosomes from the circulation was frequently controlled by the inoculation of mice.

Forty-two guinea-pigs infected with Uganda nagana were used.

Eleven of these were controls and the remainder were treated either with terebene alone, or with terebene and atoxyl. Of ten guinea-pigs infected with surra, four were controls and the remainder were treated with terebene only.

The controls infected with the Uganda trypanosomes died in from 11 to 56 days, and those infected with surra in from 17 to 41 days.

Group A.—Guinea-pigs treated solely with essence of terebene.

Series 1. Guinea-pigs receiving one or more injections of terebene, the first being given at the time of infection in order to judge the effect upon the period of incubation.

Series 2. Guinea-pigs receiving a single injection of terebene when the infection was at its maximum.

In both of these series the two species of trypanosome were used.

The conclusions drawn from these experiments may be summarised as follows: The injection of the terebene at the same time as the infective inoculation prolongs the period of incubation by some days in the case of both trypanosomes. The injection of terebene when trypanosomes are most numerous in the blood causes a diminution in the number or, more frequently, a complete disappearance, commencing 24 to 48 hours after the injection. This is followed after an interval of about five days by a relapse. In spite of the severe experimental conditions, especially in the second series, the treated guinea-pigs frequently lived considerably longer than the controls.

Group B.—Guinea-pigs included in this group were inoculated intraperitoneally with the Uganda nagana strain and then treated with atoxyl and essence of terebene.

Series 1. Guinea-pigs receiving two injections of atoxyl and terebene. These were subdivided into two lots.

a. Those that received the first injection of terebene at the time of infection and the first injection of atoxyl when the trypanosomes were very numerous in the blood.

b. Those receiving simultaneous injections of terebene and atoxyl at the height of infection. As controls infected guinea-pigs treated with atoxyl and guinea-pigs which were left untreated were used.

Series 2. Guinea-pigs receiving several injections of terebene and atoxyl.

The dose of terebene used was 1 cc. in every case, and a fresh injection was only given when the abscess caused by one dose burst.

These guinea-pigs were also divided into two lots.

In the first lot the atoxyl injections were given every eight days in two doses of 0.015 and 0.02 g. at an interval of 24 hours. In the second lot the injections were given every five days, irrespective of whether the trypanosomes were present in the blood or not. The treatment was commenced two or three days after the appearance of trypanosomes in the blood. Controls similar to those of the first series were used.

The following is a summary of the conclusions drawn:

1. Atoxyl causes only a temporary disappearance of trypanosomes from the blood of guinea-pigs inoculated with the Uganda strain of nagana.
2. The injection of atoxyl in conjunction with abscess formation resulting from the injection of essence of terebene always produces a disappearance of trypanosomes from the blood which lasts for several days.

3. Repeated application of the combined treatment effects a complete disappearance of trypanosomes which lasts for a long time, but a complete cure is not effected.

4. Guinea-pigs weighing from 400-600 grammes can withstand twelve or thirteen injections, and possibly more, of atoxyl in doses from 0.015 to 0.02 g. More than 1 cc. of terebene must not be given at a dose, and the number of injections possible depends upon the emaciation produced.

The deaths of some of the treated animals before the controls may be ascribed to this.

5. Combined treatment appears to be superior to treatment with atoxyl alone. The terebene or the resulting abscess formation probably facilitates the formation of trypanotoxyl.

6. The method should be tried upon large animals in which abscess-formation does not produce so marked a debilitating effect.

(147) KOLLE (W.), HARTOCH (O.) & SCHÜRMANN (W.). (i) **Weitere Mitteilungen über chemotherapeutische Experimentalstudien bei Trypanosomeninfektionen.** [Further Chemotherapeutic Experiments in Connection with Trypanosome Infections.]—*Deut. Med. Wochenschr.* 1914. Jan. 29. Vol. 40. No. 5. pp. 212-214.

(148) (ii) **Chemotherapeutische Experimentalstudien bei Trypanosomeninfektionen.** ii. **Mitteilung.**—*Zeitschr. f. Immunitätsforsch. u. experim. Therapie.* Orig. 1914. Jan. 22. Vol. 20. No. 5. pp. 436-475.

i. The authors reaffirm their statement published previously, to the effect that compounds of pentavalent antimony are practically useless for the treatment of trypanosomiasis.

Their experiments with Trixidin (antimony trioxide) upon large animals did not yield the satisfactory results obtained with small laboratory animals. Rabbits, dogs, and monkeys were used, and the intramuscular injection of trixidin lead to abscess formation. In a few cases, however, injections into rabbits were not followed by any untoward complications.

With the object of trying to avoid this, a number of substances were added to the trixidin suspension (in oil), but without success. The distribution of the injections over the body also failed to prevent abscess formation.

They succeeded however in administering the drug intravenously in salt solution, and they were able to cure rabbits infected with dourine by one or two injections.

By a special process it was found possible to get the antimony trioxide into a sufficiently fine state of division to permit of its being used for intravenous injection. At the same the drug presented a greater effective surface.

The good results obtained with commercial antimony trioxide are improved upon by the use of the drug in a more finely divided form, but results are not yet ready for publication. An attempt was made to treat horses infected with dourine by means of intramuscular injections, but, as in the case of the rabbits, abscess formation resulted.

OSTERTAG reports to the authors that good results have been obtained with naturally infected cattle in East Africa. Information is not given as to how the treatment has been carried out in this instance. Up to the time of writing (2 months) the treated cattle had remained free from trypanosomes.

Trixidin has been found to be capable of protecting animals against infection for a period of several weeks.

The method of making a salve either with metallic antimony or with insoluble organic compounds has been used successfully. No toxic effects have been produced.

Dimethyl-phenyl-pyrazalon-antimony-trichloride salve and intramuscular injections of trixidin are said to have a marked effect upon sleeping sickness in man, but abscess formation results from the latter treatment. No experiments have as yet been carried out using the intravenous injection method.

Conclusions :—

1. Trixidin effects a cure in the case of large animals affected with chronic trypanosomiasis.

2. In cases in which intramuscular injections of trixidin lead to abscess formation, infected rabbits and guinea-pigs can be cured by intravenous injections—antimony trioxide in salt solution.

3. A lasting cure can be effected in the case of dogs suffering from chronic dourine by the injection of antimony compounds, and particularly of dimethyl-phenyl-pyrazalon-antimony-trichloride (Scheitlin).

4. The principle advocated by us (involving the use of insoluble compounds of antimony or the inunctions) can also be applied to the human subject, but at the moment abscess formation precludes the subcutaneous or intramuscular injection of antimony trioxide.

5. Antimony trioxide can be obtained in a form resembling the colloidal form by a special process, and this can be used with great success for intravenous injection into small animals.

6. Further investigation is required to see whether colloidal antimony trioxide can be used for the treatment of sleeping sickness in man, or as an adjuvant in a combined treatment.

ii. This paper contains a fuller and more detailed account of the above experiments.

(149) MOLDOVAN (J.). Ueber die Wirkungsart des Atoxyls, Salvarsans und des Menschenserums bei der experimentellen Naganainfektion. [The Nature of the Action of Atoxyl, Salvarsan, and Human Serum in Experimental Nagana.]—*Zeitschr. f. Immunitätsforsch. u. experim. Therapie*. Orig. 1914. Mar. 14. Vol. 21. No. 1/5. pp. 481-519.

The author's experiments lead him to conclude that atoxyl acts directly upon the trypanosomes, and that the body cells, phagocytes, and other defensive powers of the body do not intervene. The atoxyl is absorbed by the trypanosomes and the toxic substance is formed in their own bodies. The animal body is necessary only in so far as it renders a multiplication of the parasites possible, involves an increase in metabolism, and leads to the absorption of substances and the production of toxic materials. In vitro the parasites are indifferent owing to their low vitality.

The property of being atoxyl-fast appears to depend not only upon absorption not taking place or the atoxyl not being reduced, but also upon the insusceptibility of the trypanosomes to the toxic products. The same holds good for salvarsan and human serum.

- (150) LURZ (R.). *Versuche mit dem Trypanosomenheilmittel "Trioxidin" bei schlafkranken Menschen.* [Experiments with Trioxidin in the Treatment of Sleeping Sickness in Man.]—*Arch. f. Schiffs- u. Trop.-Hyg.* 1914. Mar. Vol. 18. No. 6. pp. 212-213.

Trioxidin is a 30 per cent. emulsion of antimony trioxide in oil, and it is credited with effecting a permanent cure in small animals infected with trypanosomes in 100 per cent. of cases.

Seven persons infected with sleeping sickness, but shewing no clinical symptoms save swelling of the glands, were put under treatment. Two intramuscular injections were given, the doses varying from 0.1 to 0.3 grammes, and the injections being given at an interval of two days. The injections were made into the buttocks.

The injections were followed by severe abscesses, fever, pain, loss of sleep and appetite, and the patients lost condition. Seven days after the second injection the abscesses were opened and a large quantity of pus and unabsorbed emulsion escaped. The abscesses healed slowly.

The glands did not decrease in size after the trioxidin injections, and though in three cases trypanosomes disappeared from the blood and the glands, they could be found in the cerebro-spinal fluid.

The author concludes that the drug is not suitable for the treatment of sleeping sickness.

- (151) DANYSZ (J.). *Essais de Chimiothérapie. Combinaisons des Sels d'Argent et des Composés arsenicaux dans le Traitement des Trypanosomiasis Expérimentales et de la Syphilis chez l'Homme.* [Chemotherapeutic Experiments. Combinations of Silver Salts and Arsenic Compounds in the Treatment of Experimental Trypanosomiasis and of Syphilis in Man.]—*Ann. Inst. Pasteur.* 1914. Mar. Vol. 28. No. 3. pp. 238-256.

The following is an abstract of that portion of the paper which deals with the author's investigations in connection with experimental trypanosomiasis.

In the majority of the experiments the trypanosome of surra was used. Control mice inoculated subcutaneously died as a rule on the fourth or fifth day.

The following compounds were used and for the sake of brevity the author uses symbols given in brackets to designate each :

Dioxydiaminoarsenobenzol chlorargent-argentate (ACA).

Dioxydiaminoarsenobenzol bromo-argentate (ABA).

Dioxydiaminoarsenobenzol iodo-argentate (AIA).

Dioxydiaminoarsenobenzol (ddab).

All the mice received a single injection of one or the other of these drugs on the third day of infection, when trypanosomes were numerous in the blood.

It was found that "ABA" was distinctly more active than "ACA" and "AIA."

The dose which will cure a mouse by injection 60 hours after infection may be placed at 0.07 mgm. The maximum therapeutic dose is 2.5 mgm. The drug is therefore three times as active as "ddab" and 35 times as active as atoxyl.

In view of this fact "ABA" was principally used in subsequent experiments.

The objects of the further experiments were to ascertain the effects of treatment undertaken at earlier and later stages of infection, the effects of repeated small doses which if given as a single dose do not prevent relapses, and to investigate the treatment of relapses.

The experiments lead the author to conclude that (1) recovery can be effected with more certainty and with smaller doses the sooner treatment is undertaken after infection; (2) the administration of a number of small doses is more effective than that of a single large dose.

The dose required to effect a cure when trypanosomes were already numerous in the blood was five or six times that required if administered during the first 24 hours after infection.

In the treatment of relapses the same facts are to be observed as in the treatment of primary infections, with the difference that there are parasites present in the body which are more or less resistant to the action of the drug. Thus the line of treatment indicated is to give several doses at fairly short intervals.

The point is emphasised that the drugs play in reality a secondary part in the destruction of the parasites, the primary part being played by the defenses of the body which are stimulated by the drugs.

Comparative experiments shewed that "ABA" is as active against *T. rhodesiense* and *T. gambiense* as against the trypanosome of surra.

Mice infected with *T. rhodesiense* have been cured by a single injection of 1 mgm. of "ABA," and by 0.04 mgm. when combined with trypanred.

Doses of 1 mgm. and even 2 mgm. of "ABA" only delayed death for a few days in the case of mice infected with *T. dimorphon*. Mice infected with this trypanosome were, however, cured by a mixture of 3 mgm. of "ACA," 2 mgm. of "ABA," and 1 mgm. of trypanred. In another instance 1 mgm. of "ABA" with 1 mgm. trypanosan effected a cure.

The drug "ACA" was found to be far more effective than "606" in the treatment of avian spirillosis.

- (152) DANYSZ (J.). *Traitement du Surra par les Composés Arsenicaux et Arséno-Argentiques. Rapports entre les Doses Tolerées et les Doses Curatives.* [The Treatment of Surra with Arsenic and Arsenic-Silver Compounds. The Relationship between the Maximum Safe Doses and the Curative Doses.]—*Bull. Soc. Path. Exot.* 1914. Mar. Vol. 7. No. 3. pp. 200-202.

Mice infected with surra can be cured by various arsenic and arsenic-silver compounds, and in particular by atoxyl, arsenophenylglycin, chlorhydrate of dioxydiaminoarsenobenzol, and the sulphate of bromoargentate dioxydiaminoarsenobenzolate.

The author carried out a number of experiments with the object of ascertaining the ratios of the maximum dose tolerated to the dose sufficient to effect cures by a single injection.

The following ratios were established :

Atoxyl 1 : 1.

Arsenophenylglycin 3 : 1.

Chlorhydrate of dioxydiaminoarsenobenzol 8 : 1.

Sulphate of bromoargentic dioxydiaminoarsenobenzolate 35 : 1.

A number of experiments have shewn that the last drug, which the author terms "88d," can be injected repeatedly without ill effect.

- (153) FROSCH (P.) & KNUTH (P.). *Steigerung der Wirkung des Salvarsans durch Combination mit Optochininum hydrochloricum und Natrium salicylicum bei künstlicher hervorgerufenen Trypanosomenkrankheit der Pferde.* [Increase in the Activity of Salvarsan in Experimental Trypanosomiasis in the Horse through Combination with Hydrochloride of Optoquinine and Sodium Salicylate.] —*Berlin. Tierärztl. Wochenschr.* 1914. Feb. 19. Vol. 30. No. 8. pp. 133-134.

The strains used were nagana 'ferox' and von PROWAZEK's strain.

These proved fatal to horses in every case within two or three months after inoculation. In the earlier experiments salvarsan was administered in increasing doses, until doses of 7 grammes were given two consecutive days, but without result. The trypanosomes were found to be arsenic-fast when they re-appeared in the blood. The use of salvarsan combined with quinine hydrochloride yielded no results.

Optoquinine is aethylhydrocuprein, and was introduced into practice by MORGENROTH, at whose suggestion the authors employed the drug.

Seven horses were under treatment. The earlier experiments were directed towards ascertaining the effective dose and the best time for the administration. Horse 4, was treated 42 days after infection, Horse 7, nine days, and Horse 6, five days. Horse 8, received protective treatment 5 days before infection, and this was repeated 35 days after infection. Subsequent experiments shewed that the doses used in these experiments were small.

Horse 4.—Trypanosomes disappeared from the circulation rapidly, and on the sixth and seventh days the treatment was repeated, trypanosomes remaining absent from the blood. Unfortunately the animal died from tetanus 17 days later.

Horse 7.—Seventeen days after treatment a relapse occurred, and in spite of energetic treatment others followed, and the animal was destroyed.

Horse 6.—Died on the 15th day after treatment from colic. Trypanosomes had disappeared from the blood.

Horse 5.—Treated on the 15th day after infection. Trypanosomes disappeared from the peripheral blood, and the animal remained free from trypanosomes for four months. The cure was not complete, as relapses occurred which energetic treatment failed to arrest.

Fresh experiments were undertaken with two further animals, Nos. 9 and 11.

In these cases the doses were greatly increased and the method modified. Horse 11, received a single treatment, and No. 9 was treated twice with an interval of three days.

No. 11 was treated on the 6th day after infection and No. 9 on the 7th day. Up to the time of writing (2½ months) the animals had

remained free from trypanosomes and inoculation of their blood into mice has failed to cause infection.

A fuller report was to be made at the Congress of Tropical Medicine at Berlin in April.

- (154) WALKER (G. K.). **The Arsenical Treatment of Surra in Horses ; Records of Four Cases.**—*Jl. Comp. Path. & Therap.* 1914. Mar. Vol. 27. No. 1. pp. 71-75.

The treatment followed was that described by HOLMES in 1909 in the *Journal of Tropical Veterinary Science*. The animals treated were experimentally infected, and the control animal from which the treated animals were inoculated, and which had been inoculated from an infected dog, died on the 34th day after trypanosomes first appeared in the blood. Death occurred rather sooner than was expected, but the animal was old and weak.

Detailed descriptions of the courses of treatment given in the four cases are described. In general, the treatment consisted in a sub-cutaneous injection of atoxyl of 100 cc. of a 4 per cent. solution, followed by the administration *per os* of from 1.5 to 2.5 gm. of arsenious acid given in bolus. There was an interval of a day between the doses. In the first case the treatment was not well tolerated and the animal undoubtedly died from arsenical poisoning. The other three cases received full sub-toxic doses, but the trypanosomes were not destroyed, and reappeared in each case after two courses of treatment.

In the first case trypanosomes appeared in the blood during the first course of treatment only.

- (155) UHLENHUTH (P.) & SEYDERHELM (R.). **Experimentelle Untersuchungen über den Einfluss elektrischer Schwachströme auf Trypanosomen *in vitro* und *in vivo*.** [Investigations of the Effect of Electric Currents upon Trypanosomes *in vitro* and *in vivo*.]—*Zeitschr. f. Immunitätsforsch. u. experim. Therapie.* Orig. 1914. Mar. 14. Vol. 21. No. 1/5. pp. 366-377.

A suspension of blood containing *T. equiperdum* was made in salt solution and a current of 15 milliampères was passed through it. The trypanosomes were killed in 10-20 minutes. Before death occurred, the trypanosomes shewed an exaggerated motility. *T. lewisi* was found to offer more resistance to the effect of the electricity than the strain of *T. equiperdum* with which the experiments were carried out. A sub-lethal quantity of electricity reduced the infectivity of the trypanosomes.

Trypanosomes which had been destroyed by the electric current were found to be toxic for mice.

In their experiments with infected animals the authors ascertained the following facts :

If a tourniquet be applied to the leg of an infected rat and the current be passed through the limb, all the trypanosomes are killed. The application of the non-polarisable electrodes for 15 minutes daily to the abdomen and back of a mouse having trypanosomes in its blood caused an almost complete disappearance of the trypanosomes, but after the cessation of the application of the current trypanosomes returned and the infection followed a normal course.

- (156) HENNINGFELD (Fr.). Ueber die Isolierung einzelner Trypanosomen. [The Isolation of Single Trypanosomes.]—*Centralbl. f. Bakt.* 1. Abt. Orig. 1914. Mar. 21. Vol. 73. No. 3. pp. 228-240.

The author found LINDNER's method of isolating yeasts impracticable for the isolation of trypanosomes, but succeeded with the capillary tube method of FROSCHE. The tubes used had a lumen of .018 mm. and the glass was .006 mm. in thickness. A number of different liquids,—broth, salt solution, and the sera of different animals were used to dilute the blood containing the trypanosomes. This diluted blood was drawn into the tube. The tube was examined with an objective of either low or moderate power and a high eyepiece. When a trypanosome was found an object marker provided with a grease pencil was used to mark the portion of the tube containing it. The tube was then cut so as to obtain this portion. Some practice was necessary to acquire sufficient dexterity and speed.

Working with *T. brucei* and *T. equiperdum*, the author found that the parasites retained their vitality longer when the capillary tube method was used than when LINDNER's method was employed. The proportion varying from $1\frac{1}{2}$ to 3 times. Trypanosomes retained their vitality best when serum was used as the diluting fluid. In spite of the fact that in serum dilutions in capillary tubes the trypanosomes retained their vitality for as long as 23 hours, no multiplication of the parasite was observed in any instance.

A few control experiments shewed that when no moving trypanosomes were discoverable in the diluted blood the mixture was not infective for experimental animals.

In three experiments the placing of the fragment of capillary tube containing a single trypanosome under the skin of mice failed to set up infection.

OEHLER's method of drawing the fragment of capillary tube into a syringe containing 1 cc. of salt solution was also tried on two occasions, but failure to infect resulted.

The author found it an advantage to use serum as a diluent and to have serum in the syringe for injection of the tube into the abdomen. He also found it an improvement in the method to push the piece of tube into the needle (dry) of the syringe instead of sucking it up with a small drop of liquid. Working in this way the author was able to obtain 70 per cent. of successes.

- (157) LUNDIE (A.). The Detection of Trypanosomes in Animals.—*Jl. Trop. Med. & Hyg.* 1914. Jan. 15. Vol. 17. No. 2. p. 22.

The author has found the following method to yield good results where plenty of blood is available.

Tubes containing 5 grammes of potassium citrate dissolved in 5 cc. of sterile water are prepared and sterilised. The blood is allowed to run direct into the tubes, which are then set aside in the vertical position. Within half an hour there is always a little clear liquid at the top, and if trypanosomes are present in the blood at all, they will be found there.

The method is said to yield a larger number of positive results than any of the other methods known to the author.

Another advantage claimed for the method is, that it can be used for the study of trypanosomes. The tubes are left until the corpuscles have settled (three days) and the clear liquid is pipetted into a fresh sterile tube. The author has frequently obtained crithidial forms in this way.

It is also said that the life history of trypanosomes in tsetse flies can be studied by imitating the conditions present in the stomach of the fly. This is done by adding hydrochloric acid to make 0.2 per cent. strength. The effect is to convert the blood into a brown jelly. "The addition of a digestive ferment would no doubt complete the culture medium quite successfully. The matter of temperature is simple as the fly cannot possibly incubate its undigested blood at a temperature higher than that of the air. Experiments in this direction promise a certain amount of success and are certainly different from what have hitherto been in vogue."

(158) MARULLAZ (M.). *Contribution à l'Étude des Trypanosomes des Oiseaux, Deux Espèces Nouvelles.* [The Trypanosomes of Birds. Two New Species.]—*Bull. Soc. Path. Exot.* 1914. Feb. Vol. 7. No. 2. pp. 115-117. With 2 text-figs.

Trypanosoma lagonostictae. This parasite was found in smears from the liver and lungs of *Lagonosticta senegala*. The parasites were not seen in the fresh blood, the bird dying two days after arrival at the laboratory in Paris. Only one type of trypanosome was found. The parasites are somewhat stumpy, measuring 22 to 25 microns in length, including the free flagellum of 2 to 3 microns, and 5 to 7 microns in width. The body terminated posteriorly in a sharp point. The nucleus measured about 3 microns. The centrosome is very distinct and the cytoplasm slightly granular. In the anterior portion the granules form four or five very distinct rows converging towards the anterior end of the body. The trypanosomes were very scanty in the preparations from the lungs and liver, and none were found in the spleen.

Trypanosoma liothricis. A single trypanosome of this species was found in the blood of the bird during life, and none were found in films from the organs and bone marrow after death. Cultures were made from the heart blood and trypanosomes developed rapidly. In the cultures small forms measuring 21 by 4.5 microns, intermediate forms measuring 30 by 3, large stumpy forms measuring 34 by 6, and long slender forms measuring 42 by 2 microns were found. Large refractile granules could be seen in the cytoplasm in fresh preparations, but these disappeared after fixation with alcohol and ether, and were replaced by vacuoles in specimens stained with Giemsa.

In stained preparations variable numbers of chromatophile granules could be found, particularly in the anterior portion of the body. The nucleus and centrosome stained well. Crithidial forms were by far the most numerous.

Inoculation of four other birds of the same species failed to set up infection.

LEISHMANIASIS.

- (159) WENYON (C. M.). **The Culture of Leishmania from the Finger-Blood of a Case of Indian Kala Azar, with some Remarks on the Nature of Certain Granular Bodies recently described in this Disease.**—*Jl. Trop. Med. & Hyg.* 1914. Feb. 16. Vol. 15. No. 4. pp. 49-51.

Six tubes of NNN medium were inoculated with 2 to 3 drops of blood taken from the finger of a patient in whom leishmania had been found by liver puncture.

Repeated examinations of the blood had failed to reveal the parasite. On the 6th and 11th days the tubes were negative (one was contaminated), but on the 18th day flagellated forms were found in the other five.

This demonstrates the possibility of the invertebrate host becoming infected from peripheral blood. The author states that it is now a fairly common experience in animal experiments to obtain cultures from organs in which the parasite has not been detected microscopically.

The question arises whether such cultures result from some stage of the parasite hitherto unrecognised, or from leishmania which are present in numbers that are too small to permit of recognition by microscopical examination.

On a number of occasions the author has encountered the protoplasmic masses containing purple-staining granules described by ARCHIBALD, STATHAM and BUTLER, and SMALLMAN in smears obtained by liver puncture in animals. As a control to these bodies, uninoculated animals were examined by the same procedure and exactly similar bodies were found, thus doing away with any view that they may be connected with leishmania. The author is convinced that the bodies are merely detached portions of cytoplasm from large cells which are themselves charged with granules.

- (160) MAYER (M.) & WERNER (H.). **Kultur des Kala-Azar-Erregers (*Leishmania donovani*) aus dem peripherischen Blut des Menschen.** [The Cultivation of the Cause of Kala Azar (*Leishmania donovani*) from the Peripheral Blood of the Human Subject.]—*Deut. Med. Wochenschr.* 1914. Jan. 8. Vol. 40. No. 2. pp. 67-68.

The authors found parasites in tubes of NNN medium a month after they were inoculated. About a dozen tubes were used, eight of which became contaminated; but the remaining five yielded positive results. Subcultivation was also successful.

The examination of 12 smears of the blood used as seed material failed to reveal the parasite. From three to five drops of blood were used for the inoculation of each tube. This the authors calculate would have been sufficient for from 36 to 60 film preparations.

A further series of culture tubes inoculated about a month after the first failed to develop any growth.

- (161) PRINGAULT (E.). **Existence de la Leishmaniose canine à Marseille.** [The Existence of Canine Leishmaniasis at Marseilles.]—*Bull. Soc. Path. Exot.* 1914. Jan. Vol. 7. No. 1: pp. 41-42.

The author has discovered one case out of 50 dogs examined. The

animal was in very poor condition, but it was not possible to make a post-mortem. The method of examination was trephining the femur. Parasites were numerous in the marrow.

- (162) LAVERAN (A.). **Au Sujet d'un Cas de Leishmaniose canine signalé à Marseille.** [Regarding the Case of Canine Leishmaniasis reported from Marseilles.]—*Bull. Soc. Path. Exot.* 1914. Mar. Vol. 7. No. 3. pp. 173-174.

Laveran points out that as Marseilles is in daily communication with places in which canine leishmaniasis is known to occur, and since nothing is known regarding the history of the dog, it is quite likely that the animal contracted the infection elsewhere. He thinks that it would be a valuable protective measure to prohibit the importation of dogs from countries where either the canine or infantile form of the disease exists.

- (163) YAKIMOFF (W. L.) & SCHOKHOR (N. I.). i. **Recherches sur les Maladies tropicales humaines et animales au Turkestan. (1) Répartition de la Leishmaniose canine au Turkestan.** [Investigations regarding Human and Animal Tropical Diseases in Turkestan. (1) The Distribution of Canine Leishmaniasis.]—*Bull. Soc. Path. Exot.* 1914. Mar. Vol. 7. No. 3. p. 185.

- (164) ii. **Recherches sur les Maladies tropicales humaines et animales au Turkestan. (2) La Leishmaniose cutanée (Bouton d'Orient) spontanée du Chien du Turkestan.** [(2) Cutaneous Leishmaniasis of the Dog in Turkestan.]—*Ibid.* pp. 186-187. With 1 text fig.

i. The authors have examined 647 dogs and have found 157, or 24.26 per cent., infected. They have also encountered 31 cases in man (27 in children and 4 in adults).

ii. After mentioning the previous publications regarding the occurrence of this condition in natural circumstances, and its experimental production, the authors briefly describe a case which came under their own observation. The dog was in a very poor condition, had lost a great part of its coat, and had a purulent discharge from its eyes. On its back there was an ulcer measuring 7 by 5 mm. (microns according to the text), and a slightly larger one on the right side of the neck. The bases of the ulcers were covered with red granulations, in preparations from which, stained with Giemsa, typical parasites were found. Several were very large, measuring 7.85 by 2.35 microns.

The authors recall the fact that the parasites present in cases of oriental boil in Turkestan are according to their own observations: *Leishmania tropica* var. *minor* (Bukhara) measuring 3.92 by 3.14 microns, and *L. tropica* var. *major* (Termese), 5.49 by 3.92 microns.

They suggest the name *L. tropica* var. *canina* for the sub-species.

- (165) LEMAIRE (G.), SERGENT (Ed.), & LHÉRITIER. **Spécificité de la Kératite observée chez les Chiens atteints de Leishmaniose naturelle.** [The Specific Nature of the Keratitis observed in Natural Cases of Leishmaniasis in the Dog.]—*Bull. Soc. Path. Exot.* 1914. Mar. Vol. 7. No. 3. pp. 193-196.

The authors attach great importance to the keratitis observed in

natural cases of leishmaniasis, and histological examination of the cornea from an infected dog has revealed an interstitial keratitis associated with the presence of parasites. The following alterations were found in the invaded cornea: The anterior epithelium may shew slight thickenings and desquamation at places, but there is no actual ulceration. Immediately beneath the epithelium there are masses of cells and newly formed blood-vessels.

At first sight it appears as if the cornea were composed of stratified layers separated from each other by one or more layers of cells, or by a richly cellular exudate.

The exudate is most abundant in the middle portion of the cornea, where the laminae are separated off into fine fibrils. Lymphocytes containing parasites are to be found in the cellular groups.

The posterior membrane of the cornea is clearly visible. Descemet's membrane is thickened and irregular owing to the presence of rounded cells and large cells containing parasites. With the increasing duration of the lesion the exudate disappears and the corneal laminae undergo a process of fibrosis.

The authors have not been able to find any evidence of secondary infection, and they are convinced that the lesion is specific.

This fact is of great value from the point of view of diagnosis, and even for a retrospective diagnosis, for the reason that a more or less marked opacity is left in cases where recovery takes place spontaneously.

SPIROCHAETOSIS.

- (166) VON RATZ (S.). *Spirochaetose des Geflügels*. [Avian Spirochaetosis.]—*Berlin. Teirärzt. Wochenschr.* 1914. Feb. 12. Vol. 30. No. 7. pp. 117-119.

The author records an outbreak of spirochaetosis among fowls, geese, and ducks upon a farm in Hungary. Blood from a fowl received within 48 hours after death set up an infection in one of two fowls inoculated subcutaneously. Spirochaetes appeared in the blood, but subsequently disappeared and the bird recovered.

A diseased duck was despatched alive to the Institute (Budapest), but died before arrival. Spirochaetes could be found in very small numbers in the blood, but it failed to infect fowls by inoculation. The occurrence of the disease in three species of birds simultaneously suggests that *S. gallinarum* and *S. anserina* are identical.

- (167) GONDER (R.). *Experimentelle Studien über Spironema gallinarum und Spironema recurrentis*. [Experimental Studies regarding *Spironema gallinarum* and *Spironema recurrentis*.]—*Zeitschr. f. Immunitätsforsch. u. experim. Therapie*. Orig. 1914. Mar. 14. Vol 21. No. 1/5. pp. 309-325.

In a previous paper the author shewed that *T. lewisi*, if arsenic-fast, loses that property when it passed through the natural intermediate host, the rat louse. The same does not hold good for *S. gallinarum*. A strain of fowl spirochaetes which is arsenic-fast retains that property when it is passed through other birds and back into fowls.

The author was able to compare the Soudanese and Brazilian strains of fowl spirochaetes. He found that whereas the Soudanese strain protected against the Brazilian strain the converse did not hold good.

In a few experiments the author obtained the somewhat surprising result that an immune fowl can be reinfected if the spirochaete be passed through a Java sparrow, and conversely an immune Java sparrow can be reinfected after the spirochaete has been passed through a fowl.

- (168) LAUNOY (L.) & LÉVY BRUHL (M.). **Evolution de la Spirillose chez la Poule, après Splénectomie.** [The Evolution of Spirillosis in the Fowl after Splenectomy.]—*Compt. Rend. Soc. Biol.* 1914. Feb. 27. Vol. 76. No. 7. pp. 298-299.

Splenectomy is a somewhat difficult operation to perform on the fowl owing to the deep position occupied by the organ. When it is successfully accomplished recovery is complete in from 8 to 10 days, and no disturbance of health is observed.

In a general way the disease follows its normal course in fowls deprived of the spleen, but spirilla appear in the blood sooner than in normal fowls. They may be observed as early as 18 hours after inoculation, while in controls they make their appearance in about 48 hours. Further points are, that spirilla are constantly more numerous in the blood, and two crises occur. The septicaemic stage lasts longer in animals deprived of the spleen than in control animals. It commences 24 hours earlier, and terminates 24 to 48 hours later. There is, however, no increased severity of the disease from the clinical point of view, and in some cases the symptoms of intoxication, dullness, and paralysis are less marked and even unobserved in birds from which the spleen has been removed. This is possibly explained by the view that owing to the absence of the spleen destruction of the spirilla and liberation of toxic substances does not occur to nearly so marked a degree.

Antibodies are produced as in control birds, and the same blood reactions—rapid anaemia, leucocyte variations etc.—occur as in normal fowls.

- (169) ARNHEIM (G.). **Spirochaetenuntersuchungen.** [Investigations regarding Spirochaetes.]—*Zeitschr. f. Hyg. u. Infektionskrankh.* 1914. Jan. Vol. 76. No. 3. pp. 407-441. With 2 plates.

The bulk of this article consists in a description of the author's investigations of the spirochaetes which are responsible for disease in the human subject, but a short section is devoted to the question of the relationship between spirochaetes and swine fever.

Recently KING, BAESLOCK, and HOFFMANN have described a spirochaete as being the cause of this disease. The parasite has been found by these authors in the blood and in the intestinal ulcers, and from this they have concluded that the organism and the disease are causally connected. Opposed to this view are the facts that none of the species of spirochaetes discovered up to the present are capable of passing through filters, and further that salvarsan, which is a specific

for spirochaete infections, is ineffective in swine fever. The author carried out a number of transmission experiments with several strains of the virus. Small thick spirochaetes with a few turns were present in the virus, and spirochaetes were discoverable in the blood withdrawn from the caudal artery, but they were not discoverable in the heart blood. They were always present in very small numbers. They were present in the intestine, and at places where there were no diphtheritic lesions. Spirochaetes were also obtained by cultural methods from the mesenteric glands of an infected pig, but the author came to the conclusion that they were derived from the intestine and had nothing to do with the disease.

TOXOPLASMOSIS.

- (170) SARRAILHÉ (A.). *Notes sur la Toxoplasmose Expérimentale.* [Notes on Experimental Toxoplasmosis.]—*Bull. Soc. Path. Exot.* 1914. Mar. Vol. 7. No. 3. pp. 232-240.

This paper contains the author's observations upon the resistance of *Toxoplasma gondii* outside the body when placed in sterile media and exposed to temperatures between 0° and 37° C., or under the action of various injurious physico-chemical conditions, such as heating to temperatures higher than 40° C., and the effect of distilled water. The effects produced by certain heterologous sera have been investigated in order to ascertain whether they prevent or delay the multiplication of the parasite in susceptible animals. The coefficient of infectivity of the parasite for animals by various methods of infection with very weak dilutions of the virus in physiological salt solution has also been investigated.

In connection with the first of these points it was found that incubation at 37° C. destroys the vitality of the parasite in 24 hours. Maintenance at laboratory temperature for 17 days did not destroy the infectivity of the virus.

In a previous paper the author, in conjunction with MESNIL, stated that the parasite remains living and virulent in carcasses 30 hours after death.

In a new series of experiments it was found that the serous peritoneal exudate of a mouse dead 50 hours previously and swarming with bacteria proved fatal to mice by intraperitoneal inoculation. Exudate withdrawn 80 hours after death failed to set up infection.

It having been found that 24 hours' exposure to a temperature of 37° C. killed the virus, the effect of higher temperatures for shorter periods was investigated.

A quantity of peritoneal exudate was drawn into a pipette and incubated at 45° C. Mice were inoculated at half an hour, one hour, and two hours with this material. All died, but the period elapsing between inoculation and death was increased to 8 days in the first case and 11 days in the last. Control mice died in 5 days.

As the peritoneal fluid was very viscous and contained coarse particles which might assist the parasites in resisting the temperature, pleural fluid, which was much clearer, was used.

A control mouse inoculated with unheated exudate died in 7 days, and two mice inoculated with virus heated for 2 hours at 45° C. died in 13 and 15 days. Peritoneal exudate was mixed with an equal quantity of physiological salt solution, the pipette being kept in a vertical position for an hour. It was then incubated for 2 hours at 45° C. The upper clear liquid and the turbid liquid were each inoculated into two mice. The mice inoculated with the clear liquid died in 7 and 9 days, and those inoculated with the turbid liquid in 7 and 10 days. The control mouse died on the 5th day. Heating either undiluted exudate or a 10 per cent. solution of exudate to 48·5° for two hours was sufficient to destroy the infectivity of the parasite. Similar results were obtained when the temperature was 50° and 52·5° C. In all these cases the mice which failed to become infected with the heated virus were shewn to be susceptible and to have acquired no immunity by a subsequent inoculation with fresh virus.

The following mixtures were injected into mice :

| Pure virus drops. | Physiological salt solution drops. | Distilled water drops. |
|----------------------|---------------------------------------|---------------------------|
| 1 | 10 | 0 |
| 1 | 9 | 1 |
| 1 | 7 | 3 |
| 1 | 5 | 5 |
| 1 | 3 | 7 |
| 1 | 1 | 9 |
| 1 | 0 | 10 |

The mixtures were allowed to stand for 5 minutes before being used for the intraperitoneal inoculation of the mice.

The mice inoculated with the first three mixtures and the control mouse died in 5 days. The remaining mice died in 6 days.

A mixture containing 1 drop of virus in 12 drops of distilled water allowed to stand for 15 minutes before inoculation failed to set up infection.

The addition of human serum, serum from a cynocephalus, and from a rat, to the virus failed to prevent infection.

The addition of serum from an infected dog to the virus failed to prevent infection when the mixture was inoculated into mice, even though comparatively large quantities of serum were used.

A virus diluted 1 in 100,000 in salt solution was capable of infecting mice.

A mouse inoculated into the nasal fossae with a few drops of a 1 per cent. solution of virus in physiological salt solution contracted a fatal infection, and died in 10 days.

Two drops of a 1 per 100,000 dilution instilled into each eye of a mouse failed to set up infection. Negative results followed the introduction of this dilution into the nasal fossae and vagina.

As the author has very seldom found the parasite in circulating blood, he withdrew blood from the heart of an infected mouse, mixed it with citrate solution and used doses of 1, 5, and 15 drops for the intraperitoneal inoculation of three mice. All three died, in 10, 9, and 8 days respectively.

- (171) CARINI (A.) & MACIEL (J. J.). Infections de Toxoplasmose et de Paralysie Bulbaire Infectieuse par les Muqueuses Saines. [Infections with Toxoplasmosis and Infectious Bulbar Paralysis by Way of Healthy Mucous Membranes.]—*Bull. Soc. Path. Exot.* 1914. Feb. Vol. 7. No. 2. pp. 112-114.

This paper contains a brief statement to the effect that the authors have transmitted infectious bulbar paralysis to the rabbit by placing in the conjunctival sac a few drops of blood or tissue-extract from an infected animal.

They have also been able to infect pigeons by smearing the buccal mucous membrane with material containing large numbers of toxoplasms, and also by placing similar material on the eye.

PROTOZOA.

- (172) LAVERAN (A.) & MARULLAZ (M.). Sur deux *Hemamibes* et un *Toxoplasme* du *Liothrix luteus*. [Two Haemamoebae and a Toxoplasma of *Liothrix luteus*.]—*Bull. Soc. Path. Exot.* 1914. Jan. Vol. 7. No. 1. pp. 21-25. With 1 text fig.

Of seven birds (*Liothrix luteus*) purchased in Paris three shewed haemamoebae apparently belonging to two different species; two had haematozoa of the toxoplasma type, and in one a trypanosome was found.

The smaller of the haemamoebae appeared to be of the *H. relicta* type (*Proteosoma*), and the larger of the *H. ziemanni* type.

The authors propose *H. tenuis* as the name for the former and *H. liothricis* for the larger.

The toxoplasma found closely resembles that of the sparrow and paddy, and the authors suggest the name *Toxoplasma liothricis* for it.

- (173) GALLI-VALERIO (B.). *Smithia talpae* n. sp. (*Piroplasmidae*) chez *Talpa europaea* L. [*Smithia talpae* n. sp. in the European Mole.]—*Centralbl. f. Bakt.* 1. Abt. Orig. 1914. Feb. 25. Vol. 73. No. 2. pp. 142-143. With 1 text-fig.

The parasite described in this paper was found in the blood of a mole received from Malleray (Tavannes Valley, Jura).

Parasites were scanty in smears made from the blood, but possibly this was due to the fact that the blood was coagulated. Specimens stained badly both with Leishman and Bram's azur, although they were stained for 24 hours.

All the parasites were contained in red corpuscles, and two forms were observed: 1. Long pear-shaped parasites measuring from 3 to 4½ microns in length by 1 to 2 in width. The cytoplasm stained faintly blue and the chromatin was disposed at the thicker pole. 2. Round or oval bodies situated for the most part at the periphery of the corpuscles. Single parasites only were found in the corpuscles. The chromatin formed a single small mass placed at the periphery of the parasite or was in the form of a row of minute granules. The parasites measured from 1.5 to 2 microns in diameter. No multiplication forms were observed.

- (174) BEHREND (K.). Kurze Angaben über eine nichtpathogene Amöbe aus dem Darm von *Macacus rhesus*. [A Short Description of a Non-Pathogenic Amoeba from the Intestine of *Macacus rhesus*.]—*Archiv. f. Protistenkunde*. 1914. Mar. 28. Vol. 34. No. 1. pp. 35-38. With 8 text-figs.

Only encysted forms were seen with certainty, but a few other forms were observed regarding which there was some doubt as to whether they were free or encysted.

The cysts varied from 8 to 25 microns in diameter, and they were either round or oval in shape. In some cases the cyst-wall was very thin and in others sufficiently thick as to appear distinctly double-contoured.

The best stained preparations were given by Haidenhain's iron haematoxylin, Böhmer's haematoxylin, and moist Giemsa staining.

The protoplasm presents either a vacuolar or granular appearance, the vacuoles being in some cases very large.

The nucleus contains a caryosome. Specimens containing, 1, 2, 4 and even 8 nuclei were seen.

Chromidial masses are present in the cytoplasm, and these are usually rod-like.

In those parasites which were possibly not encysted the nuclei were indistinct, and the chromidia were either small or absent.

It is impossible to identify the parasite with certainty, but the occurrence of forms containing 8 nuclei suggests that it may be *Entamoeba coli*.

- (175) LAVERAN (A.) & FRANCHINI (G.). Infezione Sperimentale dei Mammiferi Mediante Flagellati del Tubo Digerente del *Ctenocephalus canis* [The Experimental Infection of Mammals with the Flagellates from the Alimentary Canal of *Ctenocephalus canis*.]—*Pathologica*. 1914. Jan. 1. Vol. 6. No. 124. p. 1.

Five white mice inoculated intraperitoneally with the contents of infected dog fleas became infected, and nine mice inoculated, some intravenously and some intraperitoneally, with the spleen or liver pulp of a mouse in which the parasites were found also became infected.

The authors were able to transmit the infection through three passages in mice. Seven mice died. The objection that the bacteria introduced with the flagellates contributed to the death of the mice is met by the fact that the infection was transmitted by blood from mouse to mouse and the blood used proved sterile by cultivation.

The disease lasts from 24 to 123 days. There is enlargement of the spleen, but in those inoculated directly from the fleas the numerous bacteria present no doubt contributed to this. Parasites may be found in the liver, blood, spleen, and bone marrow.

They resemble leishmania, and flagellate forms are very scanty: Intracellular parasites were sometimes found.

The authors have also succeeded in infecting a white rat and a dog. The rat was inoculated from an infected mouse, and parasites appeared in the blood two months later. The dog was inoculated intraperitoneally twice, with spleen and liver pulp from infected mice, and once intravenously with blood from a heavily infected mouse. The dog died 50 days after the first inoculation as the result of a complication.

Parasites were scantily present in the liver, spleen, lymphatic glands, and bone marrow. In one smear made from the axillary gland a single flagellate form was found.

- (176) SANGIORGI (G.). *Leucocytoegregarina cuniculi* n. sp.—*Pathologica*. 1914. Jan. 15. Vol. 6. No. 125. pp. 49-50. With 1 text-fig.

The author has discovered this parasite in a domesticated rabbit. The majority of the parasites occurred in a gregarine-like form. They possessed an elongated, slightly curved body with rounded ends. When stained with Giemsa the body appeared blue and finely granular, and contained towards the wider end some little specks of chromatin. The nucleus is oval and is generally placed near the narrower pole of the parasite. The chromatin is in the form of small particles embedded in an unstained matrix. The parasite is surrounded by a definite zone corresponding to a capsule.

The parasites measure 16 to 18 microns in length, by 4 to 5 in width. They are extracellular and are found scattered among the cells in preparations made from the spleen.

More rounded forms with compact-looking nuclei have been observed. Similar forms of smaller size have been observed included within mononuclear leucocytes. No multiplication forms have been seen in preparations made from any of the organs.

RABIES.

- (177) KONRÁDI (D.). Die Vererbung der Wut. iii. Mitteilung. [The Transmission of Rabies.]—*Centralbl. f. Bakt.* 1. Abt. Orig. 1914. Mar. 30. Vol. 73. No. 4/5. pp. 287-296.

Conclusions :


1. The virus of rabies passes from the mother to the foetus, but becomes attenuated by so doing. In consequence of this rabies develops progressively later in inoculated animals.

2. There appears to be no difference in this transmission in the various species of animals. It occurs in the dog, guinea-pig and rabbit, and probably in other species also.

3. In such investigations the test inoculations must be made subdurally, and guinea-pigs but not rabbits must be used. Guinea-pigs are far more susceptible to rabies, and therefore the experiments fructify more rapidly and with greater certainty than if rabbits were used. If rabbits are employed rabies either develops very late or not at all. This point was left out of consideration by previous authors who were therefore led to false conclusions.

4. The period of observation in the case of foetuses of infected animals must be longer in those cases in which guinea-pigs are used, as the disease develops after a longer interval than when the animals are infected after birth.

5. The virus of rabies is circulating in the blood of the mother at the time when the first symptoms of the disease appear, and even earlier, and it passes the placenta weeks and even months before the death of the mother.

6. The bite of a dog is dangerous 14 days before the onset of rabies. 

- (178) MANOUÉLIAN (Y.). *Recherches Histologiques sur les Glandes Salivaires dans la Rage.* [Histological Examinations of the Salivary Glands in Rabies.]—*Ann. Inst. Pasteur.* 1914. Mar. Vol. 28. No. 3. pp. 233-237. With 2 plates.

This paper contains an account of the author's investigations of the nerve ganglia in the salivary glands of about a score of dogs affected with street rabies.

Negri bodies cannot be detected in any portion of the gland tissue itself, but they are constantly present, and in large numbers, in the cytoplasm of the nerve cells of the ganglia embedded in the interstitial tissue of the glands.

The author draws attention to the possibility of mistaking débris of gland cells and other bodies for Negri bodies. For example, polynuclear leucocytes which invade the gland tissue in considerable numbers rapidly undergo degeneration. The nucleus fragments and these fragments become acidophile, staining red with Mann's stain. Glandular cells which have undergone hyaline degeneration sometimes contain bodies which may be mistaken for Negri bodies.

The lesions described by NELIS and van GEUCHTEN as occurring in cerebro-spinal sympathetic ganglia may also be found in the ganglia in the salivary glands.

As shewing how special Negri bodies are to the nerve cells, the author states that at places on the nerve trunks which accompany the vessels in the gland, small groups of nerve cells and even single nerve cells may be found, Negri bodies being found in these cells only, and never in the gland tissue or its secretions.

The following changes are observed in the acini. The acini become invaded by polynuclears, these disintegrate and the whole of contents of the acini are taken in by macrophages. In this way acini are completely changed into masses of macrophages. The cells of the acini appear to undergo a process of degeneration due to the liberation of some substance or substances through the disintegration of the polynuclears.

At places the periacinous connective tissue is also destroyed, and in this way large groups of macrophages are formed. In the ducts similar changes are observed. Large numbers of polynuclears pass into the lumen and disintegrate, and numbers of the cells lining the ducts also undergo disintegration.

These elements are found in the saliva of rabid dogs.

- (179) CRUICKSHANK (J. A.) & WRIGHT (R. E.). *A Note on Some Experiments performed with a View to finding out the Period before Symptoms during which the Saliva of an Animal incubating Rabies is infective.*—*Indian Jl. Med. Research.* 1914. Jan. Vol. 1. No. 3. pp. 532-535.

The animals used in these experiments were inoculated subdurally with street virus. The saliva was obtained either from the mouth or by fistula from the parotid and submaxillary glands. Thick suspensions were made of the glands after death which were used for subdural and intramuscular inoculation in doses of 1 to 2 cc. The

saliva was injected intramuscularly owing to the likelihood of its setting up meningitis if injected subdurally. Negri bodies were looked for in all cases in the hippocampus and cerebellum, and in doubtful cases passage experiments were resorted to.

The details of the experiments performed are given in tabular form.

Conclusions :—

"1. It is difficult to infect rabbits and guinea-pigs either intramuscularly or subdurally with the saliva of dogs or guinea-pigs in the presymptomatic stage of rabies. The table shows that in one case the saliva of a dog was infective three days before the animal showed symptoms.

"2. Even when the symptoms of rabies have manifested themselves it is still difficult to demonstrate the infectivity of saliva experimentally. This suggests that the bites of rabid dogs, even under optimum conditions for infection, may not infect because the saliva is not virulent.

"3. Suspensions of the salivary glands themselves are much more certainly infective under experimental conditions than the saliva derived from them. This is more especially the case for the submaxillary gland.

"4. Negri bodies are not always demonstrable in the brains of experimental rabid animals, although they may become so after sub-passage."

(180) **POKSCHISCHEWSKY (N.). Ueber Methoden der Schutzimpfung gegen Tollwut. [Methods of Immunisation against Rabies.]—*Zeitschr. f. Hyg. u. Infektionskrankh.* 1914. Jan. Vol. 76. No. 3. pp. 453-468.**

The author's experiments were undertaken to ascertain whether it is possible to confer active immunity upon animals, and in particular upon dogs, by means of Pasteur's original method, and whether the intraperitoneal inoculation of the fresh brain substance of passage rabbits is capable of setting up active immunity.

The following conclusions are drawn :

1. Pasteur's original method is incapable of protecting dogs and rabbits against subdural and intramuscular inoculation with street virus.

2. A sure method of infecting must be used when the immunity of animals that have been treated is being tested. Such methods are : subdural and intramuscular inoculation. Intra-ocular inoculation is less certain. Very uncertain results are given by subcutaneous inoculation, or by the bites of infected dogs.

3. In testing immunity, in addition to the method of infection, attention must be paid to the type of virus used in the test, whether it is street or fixed virus.

4. Our experiments show that the intraperitoneal inoculation of rabbits and dogs with large doses of fresh fixed virus confers a sure active immunity against intramuscular inoculation with street virus.

5. The original fixed virus inoculated intraperitoneally confers an active immunity upon dogs and rabbits in about 50 per cent. of cases against subdural inoculation, which is the most severe method of infection.

6. The intraperitoneal method of protective inoculation appears to be a promising one for application in practice, for example, for the protection of valuable dogs against the bites of rabid dogs in infected areas. It also forms a firm basis for further investigations regarding the protective inoculation of larger animals, such as the horse and ox.

RINDERPEST.

- (181) MONTGOMERY (R. E.). **Report on Double Inoculation against Rinderpest at Lumbwa (East Africa Protectorate, February 1914.)**—MS. Report dated March 30th, 1914. Received through the Colonial Office.

Five thousand seven hundred and forty-five animals were inoculated by Montgomery and the veterinary staff within a period of three days. The total mortality was 37, this figure including all animals dying within three weeks of the inoculations. Many of the deaths were due to causes other than rinderpest. Some animals, probably seven, died from redwater transmitted by the virulent blood used for the inoculations.

A tabulated statement gives the classification of all the stock in the Lumbwa District and each owner gave the number of animals which he knew to be susceptible to rinderpest. These figures cannot be taken as absolutely accurate as one death occurred among draught oxen recorded as being immune.

Bulls. 34 of which 13 were susceptible. Two were imported pure-bred animals. No deaths.

Cows. (a) In calf. 769, of which 432 were susceptible. Reactions all normal. No deaths.

(b) Heifers in calf. 498, of which 305 were susceptible. Reactions normal and no deaths.

(c) Cows not in calf. 693, of which 243 were susceptible. Reactions normal. No deaths.

Four abortions occurred on one farm among 445 in-calf animals. A report regarding the occurrence of abortion following inoculation is to be delayed for a period of three months after the inoculation.

The percentage loss of milk during the week when the reactions were at their height was 9.09 and the following week the yield was normal again.

Work oxen. 1,381, of which 433 were susceptible. One death occurred in a batch of animals believed to be immune.

Young stock. 1,177, of which 1,148 were susceptible. The reactions in these animals were rather more severe and 20 deaths or 1.74 per cent. were recorded.

Calves. 820 were inoculated. All save 11 of these were susceptible. Distinct temperature reactions followed, but the clinical symptoms were mild. There were 16 deaths, of which several were due solely to redwater.

The total death rate was 0.6 per cent., and the death rate among the susceptible animals was 1.09 per cent.

The virulent blood which served as the primary source of infective blood for the inoculations was obtained from a Uganda control at the laboratory.

- (182) HARTLEY (P.). **On the Immune Bodies occurring in Anti-Rinderpest Serum, and on the Variations occurring in the Serum Proteins during Rinderpest and during Immunisation and Hyperimmunisation.**—*Mems. Dept. Agric. in India, Vet. Ser.* 1914. Jan. Vol. 1. No. 4. pp. 178-270.

The experiments detailed in this paper were carried out with the

object of discovering some method whereby anti-rinderpest serum could be economically prepared in a stable form.

A certain degree of success has been obtained, in that it has been found possible to prepare in a powdered form those serum proteins which are combined or associated with the immunising substance. The author points out that the study of immune bodies in rinderpest offers greater difficulties than the study of immune bodies in such sera as anti-diphtheritic and anti-tetanic sera, for the reasons that the anti-serum can only be tested against the disease and not against toxin produced outside the body, and also that guinea-pigs and small animals are not available.

It was found that rinderpest immune bodies are precipitated when anti-rinderpest serum is dialysed. The precipitation is complete when tap water is used, but prolonged dialysis (6 days) against distilled water failed to complete the precipitation.

In some cases the rinderpest prophylactic could be prepared in the form of a dry powder which when injected after emulsification conferred protection upon susceptible animals.

Dilute acids exert a deleterious effect upon the immune bodies, but alkalis (0.4 per cent.) do not.

The immune bodies are associated with the euglobulins. They are precipitated by dialysis or saturation with common salt.

They are precipitated on half saturation with ammonium sulphate and are not extracted from the precipitate by saturation with common salt.

The second portion of the paper contains experiments designed to ascertain the quantitative changes occurring in the serum proteins of animals during an attack of rinderpest. The methods used were those described by GIBSON and BANZHAF in 1910.

It was found that the protein-content of the serum of animals of the same species varies considerable. The composition of serum protein of healthy animals may remain constant for periods varying from 11 to 21 days, and is not affected by repeated small bleedings at short intervals.

Examination of serum obtained from animals at intervals during attacks of rinderpest terminating fatally shewed that certain changes occur. The total heat-coagulable protein diminishes. The variations in the absolute amount of albumin per unit volume of serum are the least pronounced. A decrease was observed in the amount of the globulin and of the proteins insoluble in saturated sodium chloride. Comparison of the amounts of albumin, globulin, and proteins insoluble in common salt before inoculation, and just before death, shows that the amount of albumin has increased, and that the globulin and proteins insoluble in salt have diminished.

The third portion of the paper is on the same lines as Part II, but it deals with sera taken during immunisation and hyper-immunisation.

In 11 out of 13 animals the following changes were observed :—

1. The total protein per unit volume increased in all cases.
2. In 10 cases a decrease in the absolute amount of albumin per unit volume was observed.
3. The absolute amount of globulin per unit volume increased in all cases.

4. The most pronounced changes were observed in the proteins insoluble in saturated salt solution. An increase was observed in all cases.

In the other two animals decreases in the globulin group and in the group insoluble in salt solution were recorded. This was probably due to the fact that during immunisation the animals passed through severe attacks of the disease. Repeated large bleedings at short intervals caused a decrease in the total protein, but the various proteins are affected differently. There is little variation in the absolute amount of albumin. Globulin decreases in all cases and a more marked diminution occurs in the proteins insoluble in salt.

In Part II it is shewn that the immune bodies are associated with the latter group, and it is suggested that the large increase in this group which is associated with successful hyper-immunisation indicates an accumulation of immune bodies.

(183) MROWKA (F.). Studien ueber die Ostasische Rinderpest. [Eastern Asiatic Rinderpest.]—*Zeitschr. f. Infektionskrankh. parasit. Krankh. u. Hyg. d. Haust.* 1914. Mar. Vol. 15. No. 2. pp. 139-156. With 3 plates.

Rinderpest entirely disappears in Eastern Asia during the hot weather, and reappears with the onset of the cold weather. Sometimes intervals of several years elapse between outbreaks. The disease runs a mild course owing to the high degree of natural resistance offered. Doubts have been expressed as to whether the disease is actually rinderpest or not, but that question has been settled by experiments with animals possessing the ordinary degree of susceptibility.

The slaughterhouse at Tsingtau has furnished ample material for clinical and pathological examinations, but owing to the difficulty associated with the experimental infection of Asiatic animals, and the cost of importing susceptible animals, extensive transmission experiments have not been possible. Clinically the disease presents features which differ greatly from those seen in Germany; they suggest merely a temporary indisposition. The diagnosis is however established by the elevation of temperature, the lesions, and the sterility of the blood as regards visible bacteria, and by its virulence.

If the animals are not slaughtered, recovery almost always follows.

If an animal be killed when the febrile stage has lasted for some days, the typical lesions are found, and partially or completely healed lesions may also be seen. The author has always found fresh and healed ulcers in cases of rinderpest, and though the latter cannot have been produced during the short febrile attack, he concludes that they must be looked upon as typical. Erosion ulcers in the mouth are also pathognomic.

Lesions of the same type were found in apparently healthy animals when slaughtered. They were found in animals born during periods when the disease did not make its reappearance, and in animals which could not have passed through an attack of the disease. They were not found so frequently when a period of about three years had elapsed without an outbreak of the disease. The authors think that

this is without doubt proof of the causal relationship between the disease and the lesions.

Histological examination of the stomach lesions has been made, but the material used was obtained from apparently healthy animals, as no case of rinderpest occurred at the time when the work was being done.

The author concludes from his examinations that the erosions are primarily due to haemorrhages into the mucous coat. The haemorrhages cause local disturbances of circulation and nutrition, and autodigestion occurs. The occurrence of a succession of haemorrhages is responsible for the production of lesions which shew evidence of having been in existence for different lengths of time, and also for their occurrence in animals that are apparently healthy. The sub-mucous coat is never involved, and the ulcerations never cause perforation.

Details are given of an imported animal which was experimentally infected with blood from a native animal. The disease ran the usual course, and the symptoms and lesions were typical.

The occurrence of similar lesions in the stomachs of new-born calves indicates that the infection may be contracted in utero, and this offers an explanation of the insusceptibility of Asiatic cattle to the disease.

Animals that have contracted the disease and recovered may remain sources of infection for long periods, and may form the starting point of fresh outbreaks. Consequently quarantine is not an effective method of controlling the disease.

In this fact also lies an objection to the simultaneous inoculation method of protecting animals.

TSETSE FLIES.

- (184) WOOSNAM (R. B.). Report on a Search for *Glossina* on the Amala (Engabei) River, Southern Masai Reserve, East African Protectorate.—*Bull. Entom. Research*. 1914. Feb. Vol. 4. No. 4. pp. 271-278. With 1 map.

Abstract of conclusions :—

The tsetse fly area on the Amala River presents several points of interest. The species of fly is the Western *Glossina fusca* which has not previously been recorded in the East African Protectorate. The fly appears to be confined to the west bank of the river, or only occasionally strays to the east bank. Natives have been living for years in the district, and their cattle graze to within 400-500 yards of bush where flies exist all the year round, but they have lost only a few animals. Several sportsmen have lost mules and trek oxen.

Two explanations are offered regarding the small losses sustained by natives :

- a. The natives may be so familiar with the distribution of the fly, which appears to be confined to definite areas, that they avoid exposing their cattle.
- b. A very small percentage of flies may be infected, either because the reservoir is limited, or some climatic influence (such as altitude—5,200 feet) may affect the receptivity (ROUBAUD) of the fly.

HELMINTHS.

- (185) RAILLIET (A.), HENRY (A.), & BAUCHE (J.). Sur les Helminthes de l'Éléphant d'Asie. I. Trématodes et Cestodes. [The Helminths of the Asiatic Elephant. I. Trematodes and Cestodes.]—*Bull. Soc. Path. Exot.* 1914. Jan. Vol. 7. No. 1. pp. 78-83. With 2 text-figs.

1. Trematodes. Four species belonging to this group have been described:—(1) *Fasciola jacksoni* Cobbold 1869—bile ducts and duodenum. (2) *Pseudodiscus hawkesi* Cobbold 1875—large intestine. (3) *Watsonius ornatus* Cobbold 1882—intestine. (4) *Pfenderius papillatus* Cobbold 1882—large intestine.

2. Cestodes. (1) *Echinococcus veterinorum* Rud. 1810. This parasite has been recorded in one instance only—by BONVINCINI in 1897. (2) *Anoplocephala manubriata* n. sp. Two specimens of this parasite were found by the authors in 1910. The parasites were apparently incomplete as detached segments and groups of segments were also found. The specimens measured 1.5 and 2.6 centimetres in length, and 1.2 and 1.6 centimetres in width. Some of the free segments found were, however, 4 centimetres wide.

The scolex is large and square measuring 5 to 7 millimetres from side to side. It is somewhat compressed from before behind. There are neither hooks nor rostrum, the four suckers being separated from each other by depressions. The suckers shew concentric markings. The vertical groove is more pronounced, and it is prolonged along the upper and lower surfaces of the worm for a distance of about 3 millimetres and terminates in a shallow longitudinal fossa.

The segments are very short, and, as is often the case with the *Anoplocephala*, they are united by the central part only, the free edges forming leaf-like projections which to a great extent cover the succeeding segments.

The genital organs are completely developed at a distance of 2 centimetres from the scolex, but the uterus does not contain any eggs at that point. The testicles are small and very numerous. They are disposed in the dorsal portion of the segment and extend across practically the whole width of the parasite. There are two sperm ducts which unite and the single duct terminates in a convoluted vesicle.

The genital pore is situated towards the anterior third of the left side of each segment and takes the form of a little capsule which is directed forwards. The vagina has a fairly large receptaculum about 3 millimetres from the pore. The uterus is transversely placed and has numerous side branches.

Eggs were not found in the segments attached to the parasite, but in the free segments the uterus appeared as a single transverse row of pouches packed with eggs. The eggs had an irregularly circular or slightly polygonal outline, and measured from 70 to 80 microns in diameter. They contained a pyriform structure enclosing an embryo measuring from 17 to 22 microns.

While the worm resembles the *Anoplocephala* in its general characters, it differs from them in the disposition of the genital glands, but the authors propose to place it in that genus provisionally.

- (186) RAILLIET (A.), HENRY (A.) & BAUCHE (J.). *Sur les Helminthes de l'Eléphant d'Asie. II. Nématodes. A.* [The Helminths of the Asiatic Elephant. II. Nematodes.]—*Bull. Soc. Path. Exot.* 1914. Feb. Vol. 7. No. 2. pp. 129-132. With 1 text-fig.

Ascaridae.—

1. *Ascaris lonchoptera* Diesing, 1851 (*Strongylus elephanti* Rud., 1819),—bile ducts and duodenum.

Strongylidae.—

The parasites found in the elephant and belonging to this family are represented by the *Bunostomeae*, *Strongyleae*, *Cylicostomeae*.

Bunostomeae.—

1. *Bathostomum sangeri* (Cobbold, 1882),—intestine.
2. *Grammocephalus clathratus* (Baird, 1868),—bile ducts and intestine. This parasite also occurs in the African elephant.
3. *Bunostomum foliatum* (Cobbold, 1882),—gastric tumours and intestine.

This parasite is classified in this genus provisionally.

Strongyleae.—

1. *Evansia renniei* (Railliet and Henry, 1913),—stomach and possibly the bile ducts.

2. *Strongylus additictus* n. sp.

General characters. The body is cylindroid and somewhat attenuated at either end. The anterior end is truncated in a slightly oblique direction. The colour is greyish, but on either side there are reddish brown longitudinal stripes. The cuticle is transversely striated. The mouth is surrounded by a ring of very small plates. The buccal cavity is subglobular measuring 500 microns in length and 450 in width at the widest part. The wall is relatively thick and the anterior edge is surrounded by a distinct collar. This is incised on either side and above and below for the passage of six cephalic papillae. At the bottom of the buccal cavity there are two subventral teeth. 160 microns in length.

The oesophagus is club-shaped and rather less than 2 mm. in length. It is cylindrical for about half its length, and measures 225 microns in diameter. Its posterior extremity shews three bilobed valves.

Male.—The male is 20 mm. in length and about 1.5 in width. The caudal pouch resembles that of the other *Strongyleae*, but owing to the disposition of the posterior rays it resembles that of *Ankylostoma* rather than that of *Strongylus*. The rays are formed of a very long common trunk with poorly developed free portions.

The spicules are rather more than 2 millimetres in length, stout at their base, and taper off in a very marked manner.

Female.—Measures from 18 to 24 millimetres in length and 1.4 to 1.6 in width in front of the vulva, and 1.3 to 1.4 behind it. There is a sudden constriction at the vulva which is situated $\frac{3}{5}$ of the length from the anterior extremity. The vagina is directed forwards, but is soon lost in a convoluted mass of genital tubes. The ovijectors have not been observed. The eggs are ellipsoid, thin-shelled, and at the time of deposition segmentation of the contents is at a very early stage. The eggs measure 65-74 by 42-45 microns. The caudal extremity is in the form of a very obtuse cone and the anus opens 300 microns in front of it.

At two places in this paper the name of this parasite is given as *S. additictus*.

- (187) RAILLIET (A.), HENRY (A.), & BAUCHE (J.). *Sur les Helminthes de l'Éléphant d'Asie. III. Nématodes. B.* [The Helminths of the Asiatic Elephant. III. Nematodes. B.].—*Bull. Soc. Path. Exot.* 1914. Mar. Vol. 7. No. 3. pp. 206-210.

Stongylidae.—

Cylicostomeae. This sub-family is represented by a number of forms which present such marked differences that no doubt special genera will have to be made for them.

For the present, the authors include all, with the exception of one, in the genus *Cylicostomum* Looss (sensu lato).

1. *Choniangium* nov. gen. *Cylicostomeae* with the anterior extremity obliquely truncated so that the mouth is directed forwards and upwards. Buccal capsule larger in front than behind where the wall is thickened. There is a dorsal tunnel; no internal teeth, but irregular bosselations. Mouth surrounded by a ring of long narrow laminae. Posterior rays of the caudal pouch quadridigitate, the posterior ray being united to that of the opposite side to form a common trunk; the two intermediate rays united, and the external one separate and longer. Spicules long, slender, tubular, and tapering to the extremities which are curved. One accessory piece. Vulva close to the anus. Eggs segmented at the time of oviposition.

Type species: *Sclerostomum epistomum* Piana and Stazzi, 1900.

(1) *Choniangium epistomum* (Piana and Stazzi, 1900).—colon.

(2) *Cylicostomum sipunculiforme* (Baird, 1859).—intestine.

(3) *Cylicostomum falciferum* (Cobbold, 1882).—intestine and stomach.

This species has also been found in the African elephant.

(4) *Cylicostomum pileatum* n. sp.

General characters. Body cylindroid tapering to both extremities. Cuticle shews coarse transverse striations. At the posterior part of the body the cuticle shews some swellings having wide striations, which must be viewed as a system of secondary markings, and between which a very delicate striation can be seen. The mouth is slightly elliptical in the dorso-ventral direction. The terminal portions of the laminae (36) surrounding it form teeth about 10 microns long.

The buccal cavity is 40 microns deep. The anterior edge has an inner ring of denticulations, equal in number to those in the outer ring. At the bottom there is a third ring of very delicate denticulations. The oesophagus is formed of two subspherical swellings, the posterior of which is slightly the larger. The inner surface is very chitinous and the lumen dilates anteriorly in a funnel-like manner, effecting a wide communication with the buccal capsule. The outer surface of the whole of the anterior dilatation and a small part of the outer wall of the posterior dilatation is covered with a striated capsule 16 microns in thickness.

The wall of the intestine, particularly in the anterior part, is deeply pigmented black.

Male. The authors have had the opportunity of examining a single specimen only. This measured 9.5 mm. in length by 400 microns in width. It was characterised particularly by a long tongue-like appendage representing the posterior lobe of the caudal pouch. Just in front of the bursa there was a vesicular swelling of the cuticle. The spicules were about 2.7 mm. in length, but accurate measurement was impossible owing to the lack of material.

Female. Measures from 11·5 to 14 mm. in length, by 550 to 630 microns in thickness. The caudal extremity is conical and has the cuticular swellings already mentioned. The anus and vulva open 450–500 microns and 700–725 microns respectively from the posterior extremity. The vulva is a transverse aperture 60 microns in length. The vagina is very long and directed forwards. It divides into two parallel ovijectors in the posterior fourth of the body. The eggs are ellipsoid and are provided with thin shells. They measure 67 to 73 microns by 42 to 45 microns, and are deposited in the hollow morula stage.

Spiruridae—

Only one species appears to belong to this family.

Spiroptera smithi (Cobbold, 1882.)—tumours in the stomach with orifices communicating with the interior of the organ.

The authors think that this parasite should be placed in a genus between the *Spirura* and *Acuaria*.

Filariidae—

Only one parasite belonging to this family has been described, namely a microfilaria found by EVANS and RENNIE in the peripheral circulation.

(188) i. YAKIMOFF (W. L.) & SCHOKHOR (N. I.). *Recherches sur les Maladies tropicales humaines et animales au Turkestan.*

(4.) *Les Microfilaires des Animaux domestiques au Turkestan.*

[Investigations regarding Human and Animal Tropical Diseases in Turkestan. (4.) The Microfilariae of the Domesticated Animals in Turkestan.—*Bull. Soc. Path. Exot.* 1914. Mar. Vol. 7. No. 3. pp. 188-189.

(189) ii. YAKIMOFF (W. L.), SCHOKHOR (N. I.), KOSELKINE (P. M.), WINOGRADOFF (W. W.) & DEMIDOFF (A. P.). *Recherches sur les Maladies tropicales humaines et animales au Turkestan.*

(5.) *La Microfilariose des Chevaux au Turkestan.* [(5.) Equine Microfilariasis in Turkestan.]—*Ibid.* pp. 189-192.

i. The authors have examined camels, donkeys, mules, cattle, dogs, sheep, and goats for microfilariae, and have found them in the first four species. In four different districts the percentage of camels infected varied from 4 to 22·5. The percentage in the case of donkeys varied from 2 to 3·8. Infected mules were found in one district only, where 2 per cent. were found. Infected cattle were found in two districts, the percentage of infected animals being 5·6 and 15·2. The number of animals examined at each place is not given.

ii. Microfilariae were found in the blood of 41 horses out of 109, which arrived at Taschkent from Termese.

An extensive examination of horses in a number of districts shewed that the disease has a wide distribution. The leucocyte formula of infected horses is as follows :

| | | | | |
|--------------------------|----|----|----|------------------------|
| Lymphocytes | .. | .. | .. | 28·6 to 48·8 per cent. |
| Large mononuclears | .. | .. | .. | 0 „ 1·4 „ |
| Transitional forms | .. | .. | .. | 1·7 „ 4·3 „ |
| Neutrophile polynuclears | .. | .. | .. | 40 „ 56·1 „ |
| Eosinophiles | .. | .. | .. | 5·1 „ 11·7 „ |
| “ Türk ” forms | .. | .. | .. | 0·5 „ |
| Myelocytes | .. | .. | .. | 0·8 „ |

The percentage of infected horses varied from 0·53 to 37·5, but in all cases except the maximum given the number was below 9 per cent.

The parasite is provided with a sheath and there are three (sometimes two) white areas in the cellular column.

The first is at the anterior extremity, is oval in shape and measures from 5·68 to 10·65 microns in length. The second, which is transversely placed and measures 1·42–7·1 microns, is situated at a point 38 to 50 microns from the anterior area.

The third, which is sometimes absent, is irregularly oval and occupies the whole width of the parasite. It is placed from 142–172 microns from the anterior extremity.

The length of the parasite including the sheath varies from 270·45 to 323·76 microns, and excluding the sheath from 158·98 to 266·96 microns. The width including the sheath is from 7·1 to 11·36 microns, and excluding it from 4·24 to 9·94 microns.

At the posterior extremity there is a free portion of the sheath measuring from 4·2 to 78·1 microns.

At the anterior extremity the sheath is also always free, and in one instance in which a measurement was made the free portion measured 21·3 microns.

Three examinations as to periodicity have been made, but none has been observed.

A donkey and a sheep were found to be insusceptible.

The intravenous injection of 4·5 gm. of salvarsan caused a slight diminution in the number of parasites. The parasite is said to correspond most closely with that described by MANDEL, but a detailed description of that parasite is wanting. The chyluria mentioned by MANDEL has never been observed in Turkestan.

In the event of the parasite proving to be a new species the name *Microfilaria Ninae Kohl-Yakimovi* is suggested.

(190) WIRTH (D.). *Filariosen bei einheimischen Pferden*. [Filariæ in Native Horses.]—*Zeitschr. f. Infektionskrankh. parasit. Krankh. u. Hyg. d. Haust.* 1914. Mar. Vol. 15. No. 2. pp. 135-138.

After referring to the fact that he has already published descriptions of four cases of equine filariasis, the author proceeds to give details of a fifth case.

The case was brought to the clinique at the Veterinary High School at Vienna, in August 1912, with a history that the animal had been dull for three months and had steadily lost its appetite. The temperature was sub-normal and one micro-filaria was found in the blood.

Two months later the animal was under observation again, but in spite of repeated observations microfilariae were not found. The hæmoglobin content of the blood was 75° on Sahli's scale; the red corpuscles numbered 9,792,000 and the white 13,760 per cmm. Differential count of the white corpuscles gave the following percentages: polynuclear neutrophiles 57, eosinophiles 8, lymphocytes 31, and mononuclears 4.

On the author's suggestion BAHNMULLER carried out systematic examinations of horses for microfilariae and found 24 infected out of 280, the horses coming from different parts of Hungary. The following year 5 out of 75 animals were found to be infected.

It is probable that the severity of the symptoms depends upon the number of parasites present in the blood. To what extent mechanical and toxic effects are responsible for the symptoms is not determined.

The identity of the parasite has not been established.

- (191) DELANOË (P.). *Au Sujet de l'Existence chez un Saurien, Agama colonorum* Dum. et Bibr., d'une Filiaire et d'une Microfilarie Sanguines. [The Occurrence of a Filaria and a Microfilaria in the Blood of a Lizard, *Agama colonorum* Dum. and Bibr.]—*Bull. Soc. Path. Exot.* 1914. Feb. Vol. 7. No. 2. pp. 121-125. With text-figs.

Sheathed microfilariae were found in the blood of one lizard out of seven examined. There appeared to be no periodicity.

In moist preparations the sheath is difficult to make out except at the extremities, but the worm can be seen executing wriggling movements within it. Empty sheaths and worms devoid of sheaths were seen.

In stained preparations the worm shewed a transverse striation. Specimens measured about 190 microns in length and 6 in width.

In blood containing adult filariae the author has discovered motionless vermicule-like structures enclosed in very distinct sheaths. At one extremity a small granule with the staining reactions of chromatin could be detected. The author thinks that these bodies are atrophied eggs which have been passed out into the circulation at the same time as the completely developed microfilariae.

A group of 8 adult female filariae was found entangled with each other in the substance of the liver, but there was no degeneration of the liver tissue.

The parasites measured 12-13 centimetres in length by 0.275 millimetres in thickness, they were milky-white in colour, and in moist preparations the uterus could be seen packed with microfilariae in different stages of development.

The mouth is terminal, unarmed, and devoid of papillae. The vagina opens about 0.5 mm. behind the mouth. The cuticle is smooth, and both extremities are obtusely pointed.

OESTRIDAE.

- (192) GEDOELST (L.). *Note sur un Nouveau Genre d'Oestride.* [Note on a New Genus of Oestruus.]—*Bull. Soc. Path. Exot.* 1914. Mar. Vol. 7. No. 3. pp. 210-212.

The author refers to the description given by BLANCHARD in 1893 of a larva obtained from *Boselaphus (Bubalis) lichtensteini*, to which BLANCHARD gave the name Kirk's larva. Reference is also made to a larva in the collection at the Natural History Museum, Paris, which was found by DECORSE in the sinus of a buffalo in the Chari District in 1903. These appeared to be practically identical with each other. The author has received some larvae from the frontal sinus of a *Bubalis major* forwarded by DRAMARD. These differ only in secondary characters from DECORSE's specimens.

In the present note the author gives the characters which shew that the larvae are a special type of the sub-family Oestrinae.

The body resembles that of *Oestruus*. The antennary segments are provided with three ocelliform points, parabuccal rings unarmed, spicules exclusively ventral, and placed along the anterior edges of the third to twelfth rings, and arranged in linear groups of four to six spines on either side of the posterior margins of the fifth to eleventh segments. Posterior stigmata of the type of those of *Oestruus*. The suture which unites the false-stigma at the inner edge of the stigmatic plate joining this obliquely at its lower fourth.

The author suggests the formation of a new genus for these parasites and distinguishes at least two species, *Kirkia blanchardi* for that originally described by BLANCHARD, and *Kirkia surcoufi* for those collected by DECORSE and DRAMARD.

- (193) ROUBAUD (E.). *Oestridentes Gastricoles et Cavicoles de l'Afrique Occidentale Française*. [The Gastric Oestridae and the Oestridae of the Sinuses of French West Africa.]—*Bull. Soc. Path. Exot.* 1914. Mar. Vol. 7. No. 3. pp. 212-215.

The oestridentes have been found as parasites in the frontal sinus of *Bubalis major* (Blyth) in West Africa.

(1) *Oestruus variolosus* (Loew). Two adults of this species, described by LOEW in 1863, have been obtained by DRAMARD from larvae derived from *Bubalis major* at Boromo.

The larvae differ from those figured but not described by KING, which were found in a buffalo on the Blue Nile, by the slightly larger areas of spines, and by the absence of intermediate ventral segments, but these differences may be due to the method of preparation or to the manner of figuring.

The larvae differ from those of *Oestruus macdonaldi* Gedoelst from *Bubalis lelewel jacksoni* in the possession of smaller creeping areas which are more regularly attenuated at the sides, and also by the absence of incurvation of the 12th segment. These differences are very marked and distinguish the oestruus of the Katanga buffalo from that of the Sudan buffalo. These two larvae belong to the same group (*Oestroides* of GEDOELST) and are characterised by the cephalic segment being provided with small spines posterior to the antennal swellings and by the absence of tubercles on the ventral surface of the segments.

When fully extended *Oestruus variolosus* measures from 18 to 23 millimetres. The ventral surface is flat and the dorsal convex with 8 intermediate areas. The cephalic segment is provided with a number of short spines arranged in rows on the dorsal surface posterior to the antennal prominences. The other segments are unarmed on the dorsal surface. There are no tubercles on the lateral surfaces. The spines increase in number on the first five segments and decrease on the last two. Two rows of irregular spines on the sides of the first post-cephalic segment: a single one on the median line; two or three rows on the second; four or five on the largest posterior sections.

The areas of creeping spines form bands which taper laterally and occupy the greater part of each segment in the middle portion of the body. The spines are in the form of scales with wide colourless bases

and sharp black tips. There are two or three rows on the median post-anal prominence.

(2) *Gedoelestia cristata*. Several adults have been raised by DRAMARD from larvae obtained from Lichtenstein's buffalo. Apparently the parasite has a wide distribution, and it is not specific for the buffalo.

(3) *Kirkia surcoufi*. This parasite has also been found in *Bubalis major* and an antelope (*Cobus defassa*, probably) by DRAMARD and BOUET. Although it has a wide distribution, it is probably not found outside Central and Western Soudan. In Eastern Soudan *Kirkia blanchardi* is no doubt the species which occurs.

CONTAGIOUS PLEURO-PNEUMONIA and PASTEURILLOSIS.

(194) WALKER (G. K.). Pleuro-pneumonia of Goats in the Kangra District, Punjab, India.—*Jl. Comp. Path. & Therap.* 1914. Mar. Vol. 27. No. 1. pp. 68-71.

After reviewing briefly the literature regarding the occurrence of pleuro-pneumonia in goats, the author gives a short description of seven natural cases of the disease and of seven experimental infections. The first goat of the series of natural cases recovered from the disease and subsequently resisted experimental infection with a virulent strain of haemorrhagic septicaemia, and also failed to become infected when nasal discharge from an affected animal was rubbed on its nostrils and mouth. In the remaining six cases cultivation and microscopic examination failed to reveal organisms of any kind.

The seven experimental goats, all of the plains breed, were inoculated with exudate or blood from natural cases, but in every instance the result was negative.

Conclusions.—The pleuro-pneumonia of goats met with in Kangra appears to be identical with the enzootic pleuro-pneumonia of goats described by NOCARD and LECLAINCHE as occurring in Algeria, the Pyrenees, and the Thuringian mountains. The symptoms and lesions are similar, and the failure to transmit the disease from animal to animal is coincident.

The value of the above experiments is depreciated no doubt from the fact that plains animals had to be used. If possible, further investigations with hill goats will be made, with the object of proving the possibility of transmission by means of intrapulmonary inoculation.

(195) MITRA (S. N.). On a Peculiar Form of Pasteurella in an Indian Elephant.—*Centralbl. f. Bakt.* 1. Abt., Orig. 1914. Feb. 11. Vol. 73. No. 1. pp. 12-13. With 1 text fig.

The organism that is the subject of this note was isolated from an elephant which had been dead 36 hours before the post-mortem examination was made.

The following lesions were found at the post-mortem: There was a yellow gelatinous exudate in the tissues about the throat. The blood was thick and dark in colour. The pleural and pericardial cavities contained a blood-stained serous exudate. There was marked inflammation of the lungs, and the heart shewed a number of petechiae. The spleen was enlarged, and there were areas of extravasation under the capsule. The pulp was soft. The mesenteric glands were soft and congested. The stomach and intestines were

severely inflamed, the wall of the latter being thickened with exudate.

Microscopic examination of the various organs and fluids shewed a number of diplo-coccoid bacteria and a large number of accidental organisms. Primary cultures were not pure, but pure cultures of the coccoid organism were obtained on subcultivation. The organism was about 0.5 microns in length and stained uniformly. Its cultural characters were those of the fowl cholera bacillus.

A pigeon inoculated with the culture was dead in 8 hours, and pure cultures were obtained from the blood. Two rabbits inoculated endermically with blood from the elephant died in 18 to 24 hours. The lesions presented were those of haemorrhagic septicaemia and the blood contained large numbers of the bacteria. Pure cultures were obtained.

BOOK RECEIVED.

- (196) M'GOWAN (J. P.). [M.A., M.D., B.Sc., M.R.C.P.E.] *Investigation into the Disease of Sheep called "Scrapie" (Traberkrankheit; La Tremblante) with Especial Reference to its Association with Sarcosporidiosis. With an Appendix on a Case of John's Disease in the Sheep.*—xi + 116 pp. Demy 8vo. With 26 figs. 1914. Edinburgh: William Blackwood & Sons. (Edinburgh and East of Scotland College of Agriculture.) [Price not stated.]

[The following list of chapter headings indicates the scope of the book:—History of the disease in Britain. The disease as it occurs in other countries of Europe, and especially in Germany and France. Symptoms and pathology of the disease. Account of a case of Scrapie seen in Germany. The structure and possible developmental stages of the Sarcocyst and their relation to the spread of the disease. Symptoms exhibited by animals heavily infected with Sarcosporidia. On the action of the Sarcocystin, the toxin produced by the Sarcocyst. On the mode of spread of Sarcosporidiosis from animal to animal in carnivorous animals. Method of spread of Sarcosporidiosis in graminivorous animals. Observations as to the presence of Sarcocysts in the muscles of apparently healthy sheep. Epizootiology. Conclusions and recommendations. Appendix on a case of John's disease in the sheep. Glossary.]

RECENT LITERATURE.

[Continued from *Bulletin*, Vol. 2, No. 1, pp. 58-60.]

Babesiasis (Piroplasmosis).

- (197) NAUDIN (L.). Contribution à l'Etude de la Piroplasmose canine. [Contribution to the Study of Canine Piroplasmosis.]—*Rev. Gén. Méd. Vétér.* 1914. Jan. 1. Vol. 23. No. 265. pp. 18 to 19.
- (198) ZIEMANN (H.). Weiteres über die Zuchtung der Malaria-parasiten und der Piroplasmen (*Piroplasma canis*) *in vitro*. [Further studies on the Cultivation of the Malaria Parasites and Piroplasms (*Piroplasma canis*) *in vitro*.]—*Arch. f. Schiffs- u. Trop.-Hyg.*, 1914. Feb. Vol. 18. No. 3, pp. 77-93. With one curve.

Filariasis.

- (199) SULDEY (E. W.). Existence d'une Filaire et d'une Microfilarie chez le Caméléon de Madagascar. [The Occurrence of a *Filaria* and a *Microfilaria* in the Chameleon in Madagascar.]—*Bull. Soc. Path. Exot.*, 1914. Jan. Vol. 7. No. 1, pp. 70-71.

Leishmaniasis.

- (200) LIGNOS (A.). La Mortalité par Kala azar à Hydra pendant l'Année, 1911. [The Mortality caused by Kala azar in the Island of Hydra during 1911.]—*Bull. Soc. Path. Exot.*, 1914. Mar. Vol. 7. No. 3, p. 193.
- (201) PITTALUGA (G.). Kala azar infantile e Leishmaniosi canina in Ispagna. [Infantile Kala azar and Canine Leishmaniasis in Spain.]—*Pathologica*, 1914. Mar. 1. Vol. 6. No. 128, pp. 121-122.

Rabies.

- (202) BORELLINI (A.). Contributo allo Studio della Rabbia nei Bovini. [Contribution to the Study of Rabies in Bovines.] *Moderno Zootro. Parte Scientifica.*, 1914. Mar. 31. Vol. 25. No. 3, pp. 141-144.
- (203) KOZEWALOW (S.). Zur Virulenz des fixen Virus der Tollwut für den Menschen. [The Virulence of the Fixed Virus of Rabies for Man.]—*Centralbl. f. Bakt.* 1. Abt. Orig., 1914. Feb. 11. Vol. 73. No. 1, pp. 54-71.

Rabies, False.

- (204) VON RATZ (S.). Die Empfänglichkeit der Tiere für Paralysis bulbaris infectiosa. [The Susceptibility of Animals to Infectious Bulbar Paralysis.]—*Zeitschr. f. Infektionskrankh. parasit. Krankh. u. Hyg. d. Haust.*, 1914. Mar. Vol. 15. No. 2, pp. 99-106.

Spirochaetosis.

- (205) KING (Walter E.) & DRAKE (Raymond H.). Some Phenomena involved in the Life History of *Spirochaeta suis*. Studies on Hog Cholera.—*Jl. of Infectious Diseases*, 1914. Mar. Vol. 14. No. 2, pp. 246-250. With 1 fig.
- (206) NEUFELD (F.) & BOCKÖER (E.). Ueber die Wirkung von Salvarsan auf Hühnerspirochäten *in vivo* und *in vitro*. [The Effect of Salvarsan upon the Spirochaete of the Fowl *in vivo* and *in vitro*.]—*Zeitschr. f. Immunitätsforschung. u. experim. Therapie*. Orig., 1914. Mar. 14. Vol. 21. No. 1-5, pp. 331-341.

Trypanosomiasis.

- (207) BRUMPT (E.). Réduvidés de l'Amerique du Nord capables de transmettre le *Tryp. cruzi*. [North American Reduvids capable of transmitting *Tryp. cruzi*.]—*Bull. Soc. Path. Exot.*, 1914. Feb. Vol. 7. No. 2, pp. 132-133.
- (208) FARRANT (A. L.). Notes on Mal de Caderas.—*Veter. Record*, 1914. Mar. 28. No. 1342, pp. 626-628.
- (209) LAFONT (A.) & DUPONT (V.). Traitement de la Trypanosomiase Humaine au Sénégal par le Ludyl et le Galyl. [The Treatment of Human Trypanosomiasis in Senegal with Ludyl and Galyl.]—*Bull. Soc. Path. Exot.*, 1914. Feb. Vol. 7. No. 2, pp. 160-171.
- (210) LANFRANCHI (A.). Contributo alla Conoscenza della Forma Clinica nell'Infezione Sperimentale da Nagana, negli Equini. [The Clinical Symptoms of Nagana in Experimental Cases in Equines.]—*Clinica Veterinaria*, 1914. Mar. 15. Vol. 37. No. 5, pp. 181-190.
- (211) SCHILLING (C.). Antigene Eigenschaften verschiedener Stämme ostafrikanischer Trypanosomen. [The Antigen Properties of different Strains of East African Trypanosomes.]—*Zeitschr. f. Immunitätsforsch. u. experim. Therapie*. Orig., 1914. Mar. 14. Vol. 21. No. 1,5, pp. 358-365.

Biting Insects and Ticks.

- (212) DOANE (R. W.). Disease-bearing Insects in Samoa.—*Bull. Entomol. Research*, 1914. Feb. Vol. 4. Pt. 4, pp. 265-269. With 3 plates.

Flies.

- (213) AUSTEN (E. E.). New African Tabanidae. Part IV.—*Bull. Entomol. Research*, 1914. Feb. Vol. 4. Pt. 4, pp. 283-300. With 5 figs.
- (214) ROUBAUD (E.). Parasitisme chez les Reptiles du *Phlebotomus minutus* Rond. var. *Africanus*, Newstead. [Parasitism of *Phlebotomus minutus* Rond. var. *Africanus*, Newstead in Reptiles.]—*Bull. Soc. Path. Exot.*, 1914. Jan. Vol. 7. No. 1, pp. 83-85.

Mosquitoes.

- (215) MACFIE (J. W. Scott). A Note on the Action of Common Salt on the Larvae of *Stegomyia fasciata*.—*Bull. Entomol. Research*, 1914. Feb. Vol. 4. Pt. 4, pp. 339-344.

Ticks.

- (216) CUNLIFFE (N.). *Rhipicephalus sanguineus*: Variation in Size and Structure due to Nutrition.—*Parasitology*, 1914. Jan. Vol. 6. No. 3, pp. 372-378. With 4 text figs.
- (217) ——. Observations on *Argas brumpti*, Neumann, 1907.—*Parasitology*, 1914. Jan. Vol. 6. No. 3, pp. 379-381. With 1 text fig.
- (218) HINDLE (E.) & CUNLIFFE (N.). Regeneration in *Argas persicus*.—*Parasitology*, 1914. Jan. Vol. 6. No. 3, pp. 353-371. With 4 text figs.
- (219) ROBINSON (L. E.) & DAVIDSON (J.). The Anatomy of *Argas persicus* (Oken, 1818). Part 3.—*Parasitology*, 1914. Jan. Vol. 6. No. 3, pp. 382-424. With 4 plates and 8 text figs.

Helminths.

- (220) CONOR (A.). Essais de Transmission de la Bilharziose. [Attempts to Transmit Bilharziasis].—*Bull. Soc. Path. Exot.*, 1914. Mar. Vol. 7. No. 3, pp. 202-206.
- (221) NICOLL (W.). The Trematode Parasites of North Queensland. I.—*Parasitology*, 1914. Jan. Vol. 6. No. 4, pp. 333-350. With 2 plates.
- (222) VELU. Enzootie d'Oesophagostomose bovine au Maroc. [Enzootic Bovine Oesophagostomosis in Morocco].—*Recueil Méd. Véter.*, 1914. Mar. 30. Vol. 91. No. 6, pp. 125-127.

Protozoa.

- (223) FANTHAM (H. B.) & PORTER (ANNIE). *Herpetomonas stratiomyiae*, n. sp., a Flagellate Parasite of the Flies, *Stratiomyia chameleon* and *S. potamida*, with remarks on the Biology of the Hosts.—*Ann. Trop. Med. & Parasit.*, 1913. Dec. 30. Vol. 7. No. 4, pp. 609-620. With 1 plate.
- (224) LEGER (L.) & DUBOSCQ (O.). Sur une Nouvelle Schizogregarine à Stades Epidermiques et à Spores Monozoïques. [A New Schizogregarine with Epidermal Stages and Monozoic Spores].—*Compt. Rend. Soc. Biol.*, 1914. Feb. 27. Vol. 76. No. 7, pp. 296-297.

Unclassed.

- (225) GOZONY (L.). Die Abderhaldensche Reaktion bei protozoischer und metazoischer Parasiteninfektion. [Abderhalden's Reactions in Protozoal and Metazoal Parasitic Infections].—*Centralbl. f. Bakt.* 1. Abt. Orig., 1914. Mar. 30. Vol. 73. No. 4/5, pp. 345-349.
- (226) LUCET (Adrien). Recherches sur l'Evolution de l'*Hypoderma bovis* (de Geer) et les Moyens de la Détruire.—*Compt. Rend. Acad. Sciences*, 1914. Mar. 16. Vol. 158. No. 11, pp. 812-814.

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BABESIASIS (PIROPLASMOSIS) AND ANAPLASMOSIS.

- (228) STOCKMAN (S.) & WRAGG (W. G.). Cross Immunisation with *Piroplasma bigeminum* and *Piroplasma divergens*.—*Jl. Comp. Path. and Therap.* 1914. June. Vol. 27. No. 2. pp. 151-155.

This paper contains additional proof of the non-identity of *P. divergens* and *P. bigeminum*, in the form of cross-immunity tests.

In the first experiment detailed in the paper two heifers were used. One of these had been experimentally infected with the South African strain of red-water (*P. bigeminum*) a year previously and the other had never been exposed to infection. Both animals reacted to *P. divergens* to about the same degree, and in the blood of each parasites of the *divergens* type were found from the 3rd to the 6th and 7th days respectively.

The second experiment was carried out with an animal which had passed through a reaction due to *P. bigeminum* (African strain) and six months later had resisted a second inoculation with the same strain. After a further period of six months the animal was inoculated with blood containing *P. divergens*. A slight reaction followed, unaccompanied by any rise of temperature. Parasites of the *divergens* type were found in the blood on the 15th day, and had probably been present earlier, but had not been discovered on account of their small numbers. The absence of a rise of temperature is not significant, as the authors stated in their original communication regarding *P. divergens* that very few animals show a marked reaction to the parasite.

Experiment III. In this experiment an animal which had been infected experimentally with *P. bigeminum* two years previously became infected when inoculated with blood containing *P. divergens*.

In Experiment IV the inoculations were reversed. The animal was first inoculated with *P. divergens*, and seven weeks later with *P. bigeminum*. The experiments were successful in both instances, and the two parasites made their appearance in the blood.

In Experiment V the conditions of experiment IV were repeated and the result obtained was the same. The animal was first successfully inoculated with *P. divergens*, and after a period of seven weeks was infected with *P. bigeminum*.

Experiment VI. A heifer was inoculated with blood from an animal suffering from a naturally acquired infection with *P. divergens*. A positive reaction followed. Subsequently this animal was twice inoculated with blood from an animal that had been infected with *P. bigeminum*. No reaction occurred. The blood of this animal had, however, been giving irregular reactions with other animals. The heifer was later inoculated with the same *P. bigeminum* strain obtained from another animal, with the result that it became infected.

From this it would appear that the blood which twice failed to infect the susceptible heifer was only intermittently virulent, as it neither infected the heifer nor immunised it against a further inoculation with blood containing the same strain of parasite.

In none of the experiments recorded did either piroplasm immunise animals against the other.

(229) CHAMBERS (F.) & SMITH (J.). Immunisation of Imported Cattle against Northern Rhodesian Piroplasmosis and Anaplasmosis.—*Jl. Comp. Path. and Therap.* 1914. June. Vol. 27. No. 2. pp. 155-171. With 2 text-figs. and 6 tables.

The inoculations detailed in this article were carried out at the Maramba inoculation and quarantine station, Victoria Falls. The station was erected for the purpose owing to the fact that very large percentages of previous consignments of imported animals had succumbed to piroplasmosis and anaplasmosis. The station was provided with a dipping tank and every effort was made to render the enclosed land free from ticks. Very few ticks were found on the animals during the second month after their arrival and the further infestation of the station was stopped by dipping.

As it had been found by BEVAN that the Rhodesian anaplasmoses were more virulent than those occurring in Cape Colony, it was decided to immunise the animals by inoculating them first with blood obtained from Pretoria and then with blood derived from local sources.

The first batch of animals was given the second inoculation after an interval of five days only, with the result that the attacks of anaplasmosis due to the two viruses were in part contemporaneous and thus a dangerous state of affairs was produced, as there is at present no drug known which is of any value for the treatment of anaplasmosis. In the second batch of animals the period elapsing between the two inoculations was nine days, and in these animals two distinct anaplasmosis reactions were obtained.

It was found that the younger and poorer cattle were the best subjects for immunisation, the most suitable age being 10 to 15 months.

The most satisfactory method of immunising imported animals against local piroplasmosis and anaplasmosis is to inject subcutaneously 10 cc. of citrated blood containing *Babesia bigeminum* and *Anaplasma centrale*, followed fourteen days later by an injection of 4 to 6 cc. of blood, derived from local sources, containing *Babesia bigeminum* and *Anaplasma marginale*.

(230) LIGNIÈRES (J.). **L'Anaplasmosse bovine en Argentine.** [Bovine Anaplasmosis in the Argentine.]-*Centralbl. f. Bakt.* 1. Abt. Orig. 1914. May 27. Vol. 74. No. 1/2. pp. 133-162. With 5 text figs.

1. Anaplasmosis is endemic in certain parts of the north of the Argentine Republic, whence it may be accidentally carried to other areas by infected bovines, which form a veritable reservoir of the virus.

2. *Anaplasma argentinum* is of the same type as *Anaplasma marginale* of THEILER, but a comparative study of the two parasites is necessary before any conclusion regarding their identity or duality can be definitely arrived at.

3. In natural circumstances anaplasmosis does not occur as a pure infection; it is complicated by piroplasmoses—*P. bigeminum* and *P. argentinum*. It appears to be transmitted by the same tick—*Margaropus microplus*. Stomoxys does not act as an intermediary.

4. In order to gain a complete knowledge of anaplasmosis the parasite must be isolated, the pure disease must be studied, and the differential characters between *Anaplasma* and *Piroplasma* must be established. It is then easier to follow the different forms that anaplasmosis may take when associated with piroplasmosis.

5. Infection with anaplasmosis is equally successful whether the inoculation be subcutaneous, intravenous, or intramuscular.

6. *Anaplasma* is easily discoverable in the general circulation, particularly during the final crisis, that is to say when there are clinical symptoms and elevations of temperature. The parasites stain well and uniformly like chromatin by LAVERAN's or GIEMSA's methods. They are practically circular, are homogeneous, and vary little in size. They are generally placed at the periphery of the corpuscles. One, two, or three may be found in a single corpuscle, and they may also be found free in the plasma. At first scarcely one per cent. of the corpuscles may contain parasites, but in the later stages 30 per cent. or more may be invaded. The alterations characteristic of acute anaemia are observed in the blood at the same time.

7. Pure anaplasmosis is characterised by one or more irregular febrile crises. The most pronounced, which terminates in death, is accompanied by severe symptoms of acute anaemia, loss of appetite, extreme weakness, rapid emaciation, pallor of the mucous membranes, acceleration of the respirations and pulse, and the faeces are hard and rusty in colour.

It is a curious fact that, in spite of the rapidity and extent of the destruction of the blood corpuscles, the urine never appears to be coloured, a fact observed by THEILER. In the Argentine jaundice is rarely observed.

At the post-mortem the first noticeable thing is the pale colour of the blood, which, however, clots. The tissues are pale and sometimes present an appearance as if the animal had been bled. A yellow discolouration of the tissues is somewhat rarely observed. The muscles retain their bright appearance, but are perhaps a little pale in colour. The spleen is enlarged to two or three times the normal size, the liver is congested, the gall-bladder is enormously distended with thick or fluid granular bile.

The kidneys appear to be normal. There may be albumin in the urine, but no haemoglobin. The lymphatic glands, the alimentary tube, and the lungs do not ordinarily show any important alterations. The heart in a proportion of cases shows a large number of petechiae upon its surface.

8. In natural cases of anaplasmosis the symptoms and lesions of piroplasmosis are observed preceding those of anaplasmosis, and in particular haemoglobinuria and the nervous symptoms.

9. In areas where the disease is endemic indigenous animals suffer from it to a slight extent only.

On the other hand, imported animals, and particularly pure-bred adult animals, die in very large numbers, to the extent that it constitutes a serious obstacle to the improvement of the breeds. When resistance is offered to the disease, the period of convalescence is long, and the disease frequently takes a chronic course.

10. *Anaplasma* retains its vitality in the blood of recovered animals for long periods, in some cases for more than a year.

11. The first attack of anaplasmosis confers immunity.

12. *Anaplasma* does not appear to pass into the blood of the foetus.

13. Animals immunised against *P. bigeminum* and *P. argentinum* remain susceptible to *Anaplasma* and vice versa.

14. Blood that is rich in parasites retains its virulence after its temperature has been reduced to -20°C

15. Anaplasmosis is an entity quite distinct from the piroplasmoses.

16. *A priori* no conclusion can be drawn regarding the identity or duality of anaplasmata encountered in different countries. Definite information can only be based upon a comparative study of the parasites.

17. A diagnosis of anaplasmosis can only be established by the examination of blood taken during the algid period of the disease. If inoculation is resorted to the piroplasmoses must be taken into consideration.

The most useful methods of staining for the examination of blood are Giemsa and methylene blue. In the case of *P. bigeminum* the classical twin pear-shaped parasites will be found. In the case of *P. argentinum* the parasites will be very scanty, small and round, staining especially with methylene blue, or in the form of small twin pears and slightly lanceolated.

In cases of anaplasmosis the parasites will be found, sometimes in very large numbers, at the periphery of the corpuscles, and appearing as small, rounded, homogeneous bodies with the staining reactions of chromatin.

18. Prognosis is always unfavourable. Fat, well-bred, and adult animals are the most susceptible.

19. No specific is as yet known against anaplasmosis. Trypan-blue, which is effectual against *P. bigeminum*, is somewhat dangerous in anaplasmosis.

Sanitary measures must be put into force against anaplasmosis as against piroplasmosis: destruction of ticks, separation of infected from healthy areas. Immunisation will also prove to be of great value in the campaign against the disease.

20. Whatever method of immunisation be employed against either piroplasmosis or anaplasmosis, there can be no doubt that it is preferable to use successive doses of the pure viruses rather than to use blood the value of which is not exactly known. The immunity conferred by using pure viruses is reinforced to a valuable degree by the injection of blood derived from animals naturally infected in endemic areas and containing parasites of the same type as those used for the vaccination.

(231) MELLIS (M. C.). *Contribution à l'Étude du Traitement de la Piroplasmose bovine par le Trypanblau.* [The Treatment of Bovine Piroplasmosis by Means of Trypanblue.]—*Rev. Vét.* 1914. June. Vol. 39. No. 6. pp. 321-334.

Bovine piroplasmosis is responsible for very serious losses among cattle in Northern Africa. The disease makes its appearance about the beginning of the summer and is very variable in its manifestations. It is said that in some cases it terminates fatally within a few hours, while in other cases its onset is very insidious and its course very prolonged. The period of convalescence is very long "in the few animals which escape death."

The author gives details of 23 cases which have been treated with trypanblue.

The cases were treated in ordinary practice, and the diagnosis was not based upon microscopic examination of the blood, but upon the clinical features of the disease.

In 19 out of the 23 cases recovery followed, and in the four cases which terminated fatally the drug was administered subcutaneously. Subcutaneous injection of the drug renders its action slower, the effects not being observed until 24 to 36 hours later. After intravenous injection the effects are observed in 12 to 18 hours.

The author has always used pilocarpine in conjunction with the trypanblue, with the object of hastening the diffusion of the drug in the circulation and combating the constipation which is always observed in cases of bovine piroplasmosis.

None of the treated animals had relapses.

TRYPANOSOMIASIS.

(232) YORKE (W.) & BLACKLOCK (B.). *The Differentiation of the more important Mammalian Trypanosomes.*—*Ann. Trop. Med. & Parasit.* 1914. Apr. Vol. 8. No. 1. pp. 1-12. With 1 coloured plate and 1 chart.

This paper contains a review of the methods that are in use for the identification of different species of trypanosomes. The subject is dealt with under two main headings—the trypanosome as it occurs in the vertebrate host, and the trypanosome as it occurs in the invertebrate host.

Under the first of these headings the following characters are dealt with:—Motility, morphology, pathogenicity, symptomatology, and serum diagnosis.

Under the second heading the following points are considered :— Specific host, anatomical parts of the fly infected, period of incubation of trypanosomes in *Glossina*, morphological characters of trypanosomes in the tsetse.

Motility. This is influenced by so many variable factors that it cannot be considered as a valuable character in the identification of species.

Morphology. This is considered under two sub-headings: qualitative characters, and quantitative characters. The former are those recognisable by simple microscopic examination, and the latter require mathematical expression for their appreciation. Under the first sub-heading the various parts of the trypanosome are discussed and their value as distinguishing features considered.

With regard to the quantitative characters, the length and breadth, particularly the maximum and minimum length of monomorphic trypanosomes, are considered to be of considerable value. The average length of polymorphic trypanosomes is of little or no value, owing to the great variation from day to day in the type of the majority of the trypanosomes present in the blood.

The breadth of trypanosomes is of importance in some instances. *T. montgomeryi* and *T. congolense* have been distinguished from each other by the difference in their breadth; *T. caprae* differs from *T. vivax* in its heavier build.

The degree of *pathogenicity* and the range of pathogenicity are no longer valuable aids in the differentiation of species, recent work having shown that these are both liable under certain conditions to considerable variation.

Symptomatology is also of little value. A symptom which appears to be characteristic of a trypanosome infection is the formation of plaques in horses infected with dourine.

The reactions given by the various methods of *serum diagnosis* are recognised as being non-specific and therefore of no value for the differentiation of species. The authors have had no experience of cross-immunity as a means of identifying trypanosomes.

Invertebrate host. It appears to be probable that all varieties of tsetse are capable of transmitting most, if not all, of the African pathogenic trypanosomes, and that therefore the identification of the fly transmitting a trypanosome in a particular instance furnishes no evidence as to the identity of the trypanosome.

The part or parts of the fly involved in the development of trypanosomes may be of considerable importance in connection with the identification of trypanosomes. Upon these lines trypanosomes may be classified in three groups: (a) Those in which development is limited to the proboscis, (b) those in which the process commences in the gut and extends forwards to the proboscis, and (c) those in which it commences in the gut and is completed in the salivary glands.

The period of incubation in the fly is dependent upon the temperature and is therefore of no diagnostic value.

Sufficient is not yet known regarding the development of trypanosomes in flies to allow of the morphological changes presented by the parasites in the flies being of any value for the purposes of identification.

YORKE AND BLACKLOCK

| Group | Sub-Group | Trypanosome | Date and designators | Synonyms | Motility |
|-------|-----------|----------------------------|---------------------------|---|------------------------|
| I | A | 1 <i>T. brucei</i> | 1899 Plimmer & Bradford | | |
| | | 2 <i>T. evansi</i> | 1885 Steel | <i>T. soudanense</i> (1907) <i>T. venezuelense</i> (1910) | |
| | | 3 <i>T. hippicum</i> | 1910 Darling | | |
| | B | 4 <i>T. equiperdum</i> | 1901 Doflein | | |
| | C | 5 <i>T. equinum</i> | 1901 Voges | | |
| | D | 6 <i>T. vivax</i> | 1905 Ziemann | <i>T. cazalboui</i> (1906) | Very rapid, transverse |
| | | 7 <i>T. caprae</i> | 1910 Kleine | | |
| | | 8 <i>T. uniforme</i> | 1911 Bruce | | |
| | E | 9 <i>T. transvaaliense</i> | 1903 Laveran | | |
| | | 10 <i>T. theileri</i> | 1902 Laveran | | |
| | F | 11 <i>T. ingens</i> | 1909 Bruce | | |
| | | 12 <i>T. tragelaphi</i> | 1913 Kinghorn & Yorke | | |
| | G | 13 <i>T. lewisi</i> | 1881 Kent | | Very rapid, transverse |
| | H | 14 <i>T. cruzi</i> | 1909 Chagas | | Tumbling movement |
| II | | 15 <i>T. congolense</i> | 1904 Broden | <i>T. dimorphon</i> (1904) <i>T. pecorum</i> (1910) <i>T. confusum</i> (1909) | |
| | | 16 <i>T. nanum</i> | 1905 Laveran | | |
| | J | 17 <i>T. montgomeryi</i> | 1909 Laveran | | |
| | K | 18 <i>T. simiae</i> | 1912 Bruce | <i>T. ignotum</i> (1912) | |
| III | L | 19 <i>T. gambiense</i> | 1902 Dutton | | |
| | | 20 <i>T. multifforme</i> | 1912 Kinghorn & Yorke | | |
| | | 21 <i>T. nigeriense</i> | 1913 Macfie | | |
| | M | 22 <i>T. pecaui</i> | 1907 Laveran | | |
| | | 23 <i>T. rhodesiense</i> | 1910 Stephens & Fantham | | |
| | | 24 <i>T. ugandae</i> | 1913 Stephens & Blacklock | <i>T. brucei</i> (Uganda) (1909) | |
| | | 25 <i>T. equi</i> | 1913 Blacklock & Yorke | | |

- (233) BRUCE (D.), HAMERTON (A. E.), WATSON (D. P.) & Lady BRUCE. Description of a Strain of *Trypanosoma brucei* from Zululand. Part I.—Morphology.—*Proc. Roy. Soc.* 1914. June 4. Vol. 87. No. B. 598. pp. 493-510. With 3 plates.

"The object of this paper is to describe as fully as possible the morphology of this new strain of *T. brucei* from Zululand, in order to try to prove its identity with the trypanosome causing disease in man in Northern and Southern Rhodesia, Nyasaland, and German and Portuguese East Africa."

The strain described is one obtained from SHILSTON, Pietermaritzburg, who isolated it from an infected mule at Somkele, in Zululand.

Referring to what is called the 1896 strain of *T. brucei*, the authors quote passages in the original descriptions of this parasite by BRUCE, by KANTHACK, DURHAM, and BLANDFORD, and also by PLIMMER and BRADFORD, indicating that the parasite was dimorphic.

A comparison is instituted between a biometric chart drawn up by SHILSTON based upon the measurements of 400 trypanosomes of the 1913 strain and one constructed upon the measurements of 200 individuals contained in preparations made in 1896. These charts bear a remarkable resemblance to each other, but, as pointed out by the authors, too much importance must not be attached to this resemblance, as the measurements upon which they are based were not made under exactly comparable conditions. Comparison between the pathogenicity of the two trypanosomes for the horse and dog also reveals a close resemblance.

Morphology of the 1913 strain.

A table gives the measurements in microns of 1,000 trypanosomes from the monkey, dog, rat and guinea-pig, and a second table (reproduced) gives the average lengths of the parasite in these species:—

| Species of animal. | | Number of Trypanosomes measured. | Average length. | Maximum length. | Minimum length. |
|--------------------|----|----------------------------------|-----------------|-----------------|-----------------|
| Monkey | .. | 160 | 21.2 | 31.0 | 12.0 |
| Dog | .. | 260 | 21.5 | 32.0 | 16.0 |
| Guinea-pig | .. | 30(?80) | 22.9 | 35.0 | 17.0 |
| Rat | .. | 500 | 20.8 | 28.0 | 17.0 |

A chart constructed upon the measurements given by the 1,000 trypanosomes of the 1913 strain resembles some of the charts constructed upon the Nyasaland human strain, but differs from others.

A further series of charts is constructed upon the measurements of 500 individuals taken on nine consecutive days in the blood of rats. Some of these charts record the measurements after the passage of the trypanosome through *G. morsitans*, once, twice and three times. The differences between these charts are very slight. Curves are given comparing the 1913 strain of *brucei* and the Nyasaland wild game strain and a certain degree of resemblance is observed.

Conclusions:—

"1. The trypanosome described in this paper under the name of the "Zululand strain, 1913," is the same species as that discovered by Bruce in Zululand in 1894; reported on by KANTHACK, DURHAM and BLANDFORD in 1898; and named *T. brucei* by PLIMMER and BRADFORD in 1899.

"2. As regards its morphology, this trypanosome is absolutely identical with the trypanosome causing disease in man in Nyasaland, the *T. rhodesiense* of STEPHENS and FANTHAM."

Part II.—Susceptibility of Animals.—*Idem.* pp. 511-516.

The Commission had no opportunity of studying the effect produced by the "1913 strain" on the horse, but SHILSTON found that it proved fatal in 35 days. In BRUCE'S "Further Report on Nagana, in Zululand, 1896," it was recorded that two horses were killed by the "1896 strain" in 30 and 49 days respectively.

Two out of six oxen inoculated became infected. One died in 310 days, and the other apparently made a complete recovery. Four out of seven goats became infected and died. The average duration of the disease was 77·2 days. There was no oedema of the face or corneal opacity observed.

Sheep could not be procured for experiment.

Eight monkeys died in from 8 to 65 days (average 29·2).

Seventeen dogs died in from 12-26 days (average 18·5). In eight of these there was opacity of the cornea, and in two swelling of the limbs.

Eight experiments with rabbits are recorded, but six of these were reported by SHILSTON. The average duration of the disease was 32·7 days.

Ten guinea-pigs were inoculated. All became infected and died, but four of them were inoculated more than once before infection was set up. The disease lasted from 15 to 89 days (average 43·7).

Twenty-three rats were inoculated and all died, the average duration of life being 27 days.

These results are compared in tabular form with the results obtained with the strain causing disease in man in Nyasaland. The following tables are slightly abbreviated :—

Zululand strain, 1913.

| | Horse. | Ox. | Goat. | Monkey. | Dog. | Rabbit. | Guinea-pig. | White rat. |
|--------------------------|--------|-----|-------|---------|------|---------|-------------|------------|
| Average duration in days | 38 | 310 | 77 | 29 | 18 | 33 | 44 | 27 |
| Animals used | 3 | 1 | 7 | 8 | 17 | 8 | 10 | 23 |

Human strain, Nyasaland.

| | Horse. | Ox. | Goat & Sheep | Monkey. | Dog. | Rabbit. | Guinea-pig. | White rat. |
|--------------------------|--------|-----|--------------|---------|------|---------|-------------|------------|
| Average duration in days | 0 | 134 | 42 | 26 | 34 | 28 | 67 | 30 |
| Animals used | 0 | 1 | 29 | 20 | 25 | 7 | 15 | 21 |

RECOVERIES.

Zululand strain, 1913.

| | Horse. | Ox. | Goat. | Monkey. | Dog. | Rabbit. | Guinea-pig. | White rat. |
|--------------|--------|-----|-------|---------|------|---------|-------------|------------|
| Percent-ages | 0 | 83 | 0 | 0 | 0 | 0 | 0 | 0 |
| Animals used | 3 | 6 | 4 | 8 | 17 | 8 | 10 | 23 |

Human strain, Nyasaland.

| | Horse. | Ox. | Goat & Sheep | Monkey. | Dog. | Rabbit. | Guinea-pig. | White rat. |
|--------------|--------|-----|--------------|---------|------|---------|-------------|------------|
| Percent-ages | 0 | 80 | 0 | 0 | 0 | 0 | 0 | 0 |
| Animals used | 0 | 5 | 29 | 20 | 25 | 7 | 15 | 21 |

Conclusion :—

“The pathogenic action of *T. brucei*, Zululand strain, 1913, on various animals is so similar, not only in regard to the symptoms during life but also in the *post-mortem* appearances and rate of mortality, to that of the trypanosome causing disease in man in Nyasaland, that it affords another proof that these two trypanosomes are identical.”

Part III.—Development in *Glossina morsitans*.—*Idem*. pp. 526-531.
With 1 plate.

Seven experiments were made with laboratory-bred flies. Three of these were positive and four negative. Two experiments made with wild flies both had positive results.

Three hundred and eight laboratory-bred flies were used and 15·3 per cent. were found to be infected. In some cases the number of infected flies in a batch was found to be very small (1 in 58), and in others it was very high (20 in 45).

Of 100 wild flies used in the two positive experiments only 21 were dissected, and of these 5 were found to be infected.

The average period elapsing before the fly becomes infective appears to be 21 days.

Dissection of the laboratory-bred flies used in the positive experiments showed that in two the labial cavity and the hypopharynx contained trypanosomes, and that in one of these and in five others the salivary glands were infected.

None of the laboratory-bred flies used in the negative experiments had any trypanosomes either in the proboscis or in the salivary glands.

The trypanosomes found in different stages of development in the flies agree very closely in morphology with those found in flies infected with the trypanosome causing disease of man in Nyasaland.

Conclusions.

"1. *T. brucei*, Zululand, 1913, belongs to the same group as *T. gambiense* as regards its cycle of development in the tsetse fly.

"2. It has been shown that the trypanosome causing disease in man in Nyasaland also belongs to the same group.

"3. The cycle of development of the Nyasaland and Zululand trypanosomes in *G. morsitans* is so marvellously alike that it affords another reason for believing in the identity of these two trypanosomes."

(234) OEHLER (R.). Untersuchungen über den Dimorphismus von *Trypanosoma brucei*. [Investigations regarding the Dimorphism of *T. brucei*.]—*Zeitschr. f. Hyg. u. Infektionskr.* 1914. Apr. 1. Vol. 77. No. 2. pp. 356-370. With 6 text-figs.

The author applied his method of inoculation with single trypanosomes to a strain of *T. brucei* brought from East Africa by BRAUN and TEICHMANN. This strain showed marked polymorphism, and posterior nuclear forms were observed among the short aflagellar trypanosomes.

Forty inoculations into mice with single trypanosomes were made and infection resulted in two instances only. The period of incubation was 6 days in both instances, as compared with 4 to 6 days after inoculation with a large number of trypanosomes.

This proportion of successes was much lower than had been obtained previously with a virulent strain of nagana obtained from Togoland. In that case the successes had numbered 20 per cent. The author ascribes this difference to a difference of virulence in the two strains.

The infections in the two infected mice were as markedly polymorphic as the original strain, and this polymorphism was recognisable from the first day of infection.

The conclusions drawn from these facts are :

1. The dimorphism of *T. brucei* is not due to a mixed infection.
2. The dimorphism of *T. brucei* has probably nothing whatever to do with the differentiation of the sexes.

An account is given of some experiments shewing that a mixed infection may be purified by passing the trypanosomes not only through different species of animals, but even through the same species.

A mixture of a salvarsan-fast strain of the Togoland nagana strain and a salvarsan-susceptible strain of the same trypanosome was inoculated from mouse to mouse in series. By adding salvarsan solution to blood from an infected mouse the two trypanosomes could be recognised in moist preparations, as the salvarsan-fast trypanosomes retained their vitality, while the salvarsan-susceptible trypanosomes rapidly became immobilised. As the number of passages became greater the number of salvarsan-fast trypanosomes decreased, and after about 40 passages they could not be detected in the manner described. The disappearance as judged by this method was not complete, as could be proved by treating a mouse inoculated with such a strain with salvarsan in a dose of 1.0 mgm. This sufficed to kill off the susceptible trypanosomes, but the salvarsan-fast trypanosomes remained and reappeared in the blood.

In the case of antimony tartrate-fast strains the purification of a mixture occurs in the same way, but at a greater rate. The purification of mixture of the type described, and probably of other types also, is due to the difference in the rapidity of multiplication.

Regarding the significance of dimorphism, the author is of the opinion that the changes in shape of the trypanosomes are, so to speak, the anatomical expression of the remittent nature of the disease. He believes that the slender forms herald a multiplication of the trypanosomes and the short, broad forms a remission.

- (235) OEHLER (R.). **Der Dimorphismus des *Trypanosoma brucei* bei experimenteller Behandlung.** [The Dimorphism of *Trypanosoma brucei* in Experimental Treatment.]—*Zeitschr. f. Hyg. u. Infektionskr.* 1914. June 24. Vol. 78. No. 1. pp. 188-192.

The author produces evidence regarding the connection between the dimorphism of the trypanosome and the remittent nature of the infection.

Mice can be incubated at 35°C. without ill-effect provided they are given water to drink, and infection with trypanosomes does not cause the temperature to rise to a height that proves fatal (38·5°) even in mice kept in an incubator at 35°C.

Mice of the first ten passages suffered from a chronic infection, death taking place in from 20 to 70 days. And the course of the infection was marked by numerous remissions, during which the trypanosomes entirely disappeared from the blood. Broad forms present in the blood increased to a marked extent at the remissions. If such a mouse be placed in an incubator when trypanosomes are more or less numerously present in the blood, a remission with predominating broad forms will be produced in all probability in two or three days.

In mice of the 6th to 10th passage kept at room temperature the disease became acute, but in mice of the same passages kept in the incubator the disease remained chronic, relapses occurred, and dimorphism was marked.

A very similar result was observed with mice of the 20th to 30th passages.

Remissions produced by drugs also coincided with an increase in the proportion of broad forms present, but the proportion was not very high.

Inoculation with the spirillum of recurrent fever on the day following inoculation with the trypanosome also lead to a remission coincident with the increase in the number of spirilla, but in this case no broad forms were discovered.

- (236) BRUCE (D.), HAMERTON (A. E.), WATSON (D. P.) & Lady BRUCE. **The Trypanosome causing Disease in Man in Nyasaland. Part III. Development in *Glossina morsitans*.**—*Proc. Roy. Soc.* 1914. June 4. Vol. 87. No. B. 598. pp. 516-525. With 1 coloured plate.

The investigation of the development of this trypanosome in the fly was rendered difficult by the fact that it was impossible to find the pupae of the fly in any large numbers. The laboratory-bred flies had therefore to be hatched out of pupae obtained from captive flies. A further difficulty was that flies so hatched are not so strong and healthy as flies hatched out from wild pupae.

Eleven experiments were carried out with laboratory-bred flies. Three were positive and eight negative.

Five experiments were carried out with wild flies and all were positive.

Although only three of the eleven experiments carried out with laboratory-bred flies were positive, infected flies were found in eight of the experiments; the total number of infected flies (found by dissection) was 25 and the total number of flies used was 287. The percentage of infected flies was therefore 8·7. The period elapsing before infected flies became infective varied in the three positive cases from 14 to 31 days. This period was arrived at by subtracting seven days from the period elapsing between the first infected feed and the appearance of trypanosomes in the blood of the experimental animal, seven days representing the period of incubation in the experimental animal.

Attention is drawn especially to the fact that the period elapsing before the flies became infected was very short in one instance—14 days.

Two of the positive results obtained in the five experiments carried out with wild flies were evidently due to the presence of naturally infected flies, as the periods elapsing before the flies became infected were only one and three days. In the other three experiments the periods were 18, 25 and 30 days. In all, 255 flies were used, and of these 9 per cent. were found to be infected.

All flies dying during the course of the experiments were dissected. In the three positive experiments with laboratory-bred flies nine infected flies were found. In none of these were any trypanosomes found in the proboscis, labial cavity, or hypopharynx. In one or other of the flies the proventriculus, fore-, mid- and hind-gut, and salivary glands were found to contain trypanosomes, and in one of them all these parts were found to contain them.

Dissection of the flies used in the positive experiments with wild flies shewed a similar condition of affairs, but there was infection of the proboscis with trypanosomes in five cases. It is pointed out, however, that with trypanosomes numerous present in the salivary glands it would be natural to expect to find some in the proboscis, this term including the hypopharynx. It is further pointed out that in some of the wild flies the infections of the proboscis were due to *T. pecorum*, *T. simiae*, or *T. caprae*.

One of the laboratory-bred flies used in the experiments which gave negative results was found to have trypanosomes in its salivary glands, and no explanation of the failure of this fly to infect can be given. The remainder had trypanosomes in neither the proboscis nor the salivary glands.

The development of the trypanosome in the fly is said to be exactly parallel to that of *T. gambiense* in *G. palpalis*. The fly becomes infective in 14 to 31 days after the infecting meal.

The conclusions drawn are:

"1. The trypanosome causing disease in man in Nyasaland belongs to the same group as *T. gambiense*, the development taking place in the alimentary tract and salivary glands, not in the proboscis, of the fly.

"2. The percentage of flies which become infected is the same as in *T. gambiense*, 8 per cent.

"3. The percentage of flies which become infective is about 1 per cent.

"4. The length of time which elapses before a fly becomes infective varies from 14 to 31 days, average 23 days.

"5. The infective type of trypanosome in the salivary glands—corresponding to the final stage of the cycle of development—is similar to the short and stumpy form found in the blood of the vertebrate host."

- (237) YORKE (W.) & BLACKLOCK (B.) **The Identity of *T. rhodesiense* with the Trypanosome of the Same Appearance found in Game.**
—*Brit. Med. Jl.* 1914. June 6. pp. 1234-1236.

This paper is called forth by one published by KLEINE, in which the view generally held by German investigators, namely, that the trypanosome occurring in game in Rhodesia and that causing disease in man are not identical, is confirmed. The arguments used by KLEINE in support of his view are examined in detail.

The German investigators agree that the two trypanosomes are morphologically identical and that they cannot be distinguished on the grounds of pathogenicity or method of development in the fly, but they nevertheless believe the parasites to be different. KINGHORN and YORKE are said to have failed to have observed the presence of *T. brucei* in heavily infested *morsitans* areas, but it is pointed out that this assumes that the two parasites are distinguishable from each other.

The second argument used by KLEINE, namely, that the finding of infected animals in certain *morsitans* areas while no cases of sleeping sickness occurred is sufficient to separate the two parasites, is criticised because of the difficulty of ascertaining with absolute certainty that no case of human trypanosomiasis occurs in any particular district. The opponents agree that there can be no question of acquired immunity, but they disagree sharply regarding the possibility that human beings are generally very resistant and only occasionally susceptible, the English authors holding that such may be the case. In support of the English view mention is made of the recent work done in connection with the infection of rabbits with *T. vivax* and that concerning the identity of *T. nanum* and *T. congolense*.

TODD's observation is referred to, in which a European and two natives were inoculated with blood from a cow suffering from trypanosomiasis of an undetermined type, and it is pointed out that this observation has no actual bearing upon the question, for the very reason that the species of trypanosome used was not determined, and that it was probably either *T. vivax* or *T. congolense*, as these are more commonly found in cattle than the polymorphic type of trypanosome. TAUTE's experiment conducted upon himself is explicit, and whereas the German observers refer to it as supporting their view that the trypanosomes are different, the English authors consider it as affording strong support to their view that human beings are capable of offering a high degree of resistance to infection with trypanosomes.

Further support of the English view is drawn from the fact that the disease in human beings occurs sporadically in widely scattered areas, and this suggests that there must be some endemic reservoir apart from human beings.

WECK's observations regarding the susceptibility of the antelope and the possibility of infecting monkeys from the antelope are further evidence in support of the authors' view, because "to assume that the game is infected with two different trypanosomes, alike in every respect with the single exception that one variety is fatal to man whilst the other is innocuous, would be, in our opinion, excessive theorising."

Conclusions :

" In favour, then, of game being the reservoir of human trypanosomiasis in South Central Africa we have the following facts :

" 1. Human beings and game are known to be infected with trypanosomes identical as regards morphology, pathogenicity in laboratory animals, and their development in *G. morsitans*.

" 2. The human trypanosome can be successfully inoculated into game.

" 3. The peculiar sporadic occurrence of the disease in human beings suggests that they were infected from a widely spread reservoir of the infection (the game) rather than from one another.

" In conclusion, we submit that the hypothesis that man enjoys marked natural immunity, and is in consequence to a great extent resistant to infection with this parasite, affords a satisfactory explanation of the distribution of the disease, of its comparative rarity, and of the fact that TAUTE's attempt to infect himself failed."

(238) WATSON (E. A.). i. **Report on Dourine.**—*Dept. of Agriculture, Canada. Report of the Veterinary Director-General* (F. Torrance, B.A., D.V.S.) *for the Year ending 31st Mar. 1913.* Ottawa : Printed by C. H. Parmalee. pp. 81-86.

ii. **The Serum Reactions and Serum Diagnosis of Dourine.**—*Ibid.* pp. 102-107.

i. Particular interest attaches to an outbreak of the disease at Unity, Saskatchewan, which the author investigated from March to July 1912. The cases were very mild, and there was some difference of opinion as to whether they were dourine or not. Very slow progress in the disease was observed, the animals differing little in condition when the author saw them two months after illness had first been observed ; in fact, according to some who had seen the animals, there was actual improvement.

Microscopic examination for the trypanosomes failed, but a positive agglutination reaction was obtained in dilutions up to 1 in 2000, and an increased globulin content in the blood was detected by the acetic acid test (see further paper by Watson). Positive reactions were obtained with blood from other mares, and some died from dourine during the course of the investigations. In one case *T. equiperdum* was discovered.

A tabulated statement gives a brief history of nine animals which came under observation.

In the case of the stallion from which the mares became infected the infection took about a year and a half to develop into a definable malady. Experience shows that in mares the disease may be latent during pregnancy and develop rapidly after parturition, infected animals then being a source of great danger to contacts.

During the period 1906 to 1912 Watson made numerous attempts to infect dogs, rabbits, mice and other laboratory animals with *T. equiperdum*, but without success. Only partial success followed the inoculation of large numbers of rats with a strain of the trypanosome which had been passed through a series of ten foals and had acquired a high degree of virulence. Although some of the rats became infected it was found impossible to carry on the strain.

Finally, after a large number of failures, a mouse was successfully inoculated from a mare upon which successive crops of plaques made their appearance. Subsequently there was no difficulty in carrying on the strain in the laboratory.

In mice death usually occurred on the fourth day, and in rats on the fifth. The virulence of the strain for these animals has increased until rats died in three days, and mice in 48 hours.

The strain has also increased in virulence for the horse. In two instances horses inoculated from rats have shown a heavy blood invasion, and one died from dourine in six weeks. Two dogs which licked the carcass became infected and died.

ii. The following reactions are considered by Watson to be of value for the diagnosis of dourine :—

1. Complement fixation test.
2. Agglutination test.
3. Precipitin test.
4. Acetic acid test.

Complement Fixation Test. The special point about this test as applied to the diagnosis of dourine lies in the method of preparing the antigen. The blood of a number of rats at the height of infection, but before they appear to be dying, is collected. It is defibrinated with glass threads and washed five or six times, the layer containing the trypanosomes being pipetted off each time. The final emulsion of trypanosomes in salt solution is diluted one in ten with salt solution, shaken for two or three days, and then passed through a Berkefeld filter. This filtered liquid constitutes the antigen.

The amboceptor, complement, and antigen are titrated and three tubes are prepared from each sample of serum to be tested. Positive and negative controls with known sera are used in every case, together with the usual general controls.

Agglutination Test. The antigen used in this test is the emulsion described above without filtration. Special care must be exercised in making the emulsion, and it must be rejected if for any reason it shows the slightest tendency to auto-agglutination. This may arise if the blood is collected when the animal is approaching death, if it be contaminated, etc.

A number of stock dilutions of serum are prepared, and from these series of dilutions from 1 in 20 to 1 in 10,000 are made. One cubic centimetre of such dilutions is placed in a series of tubes, to each of which are added two or three drops of the trypanosome suspension. A control tube containing only salt solution is included in the series. The tubes are incubated at 37° C. for one or two hours, and the reactions are noted every half hour.

The Precipitin Test. The antigen used in this test is exactly like that used for the complement test, save that it is not diluted.

In each tube 0.55 cc. of antigen is placed, and the serum is added by means of a capillary tube which is passed through the antigen to the bottom, the serum being then allowed to escape and carry the antigen up the tube without mixing with it. Three tubes should be used in each series so that pure serum, serum diluted 1 in 5, and 1 in 10 may be used. In tubes containing diluted sera the formation of a precipitin ring marks an absolutely specific result.

The Acetic Acid Test. This test is based upon NOGUCHI's observations regarding the increased globulin-content of the serum and cerebro-spinal fluid in cases of syphilis.

Watson replaces the butyric acid used by NOGUCHI by acetic acid, after having used butyric acid in a number of tests.

One cubic centimetre of serum is placed in a graduated centrifuge tube with 9 cc. of half-saturated ammonium sulphate. The tubes are allowed to stand for an hour, and the precipitated globulin is then sedimented by centrifuging. The clear liquid is then replaced by salt solution, in which the globulin dissolves.

This solution is then distributed into tubes in the following quantities:—1.75, 1.50, 1.25, 1.00, and 0.75 cc. The volumes are then made up to 2 cc. with salt solution, and finally 1 cc. of 10 per cent. acetic acid is added to each tube. The tubes are shaken, incubated for two or three hours at body temperature, and the results read off.

Normal sera give a slight opalescence in the first tube, and perhaps a faint cloudiness in the second, but with dourine sera there is marked cloudiness which gradually increases and after 24 hours settles to the bottom as a precipitate.

If more concentrated solutions are used the reaction occurs more quickly, but the differentiation is scarcely as well defined.

In cases of dourine a positive reaction may be obtained in the early stages of the disease. A reaction has been obtained a fortnight after experimental inoculation in a horse.

Watson has carried out the test with sera derived from cases of coital exanthema, glanders, influenza, fistulous withers, and one case of swamp fever. In the last-named a positive reaction was obtained. When the serum was tested by agglutination for dourine the result was negative.

The principal value of this test, perhaps, lies in the exclusion of dourine, for in no case of dourine of known history has the test ever given a negative result.

- (239) GRAHAM (L. W.) & HUTCHISON (R. H.). **The Influence of Experimental Trypanosomiasis upon the Body Temperature of White Rats.**—*Amer. Jl. of Trop. Dis. & Prevent. Med.* 1914. May. Vol. 1. No. 11. pp. 760-775. With 3 plates comprising 7 charts.

The experiments detailed in this paper were designed to investigate the question of the variations of body temperature in the rat under normal and abnormal conditions, and also after infection with trypanosomiasis.

The records were made with thermo-electric couples, a modification of the thermopile, as the mercury thermometer is not sufficiently sensitive. The operation of the apparatus depends upon the fact that when a junction of two dissimilar metals is heated a current is generated, such current being capable of measurement. The method is said to be accurate to one-tenth of a degree centigrade.

Readings of body temperatures were made at short intervals over comparatively long periods in normal rats kept under three sets of external conditions with regard to temperature, viz., room temperature, 37° C., and refrigerator temperature.

A similar series of readings were then taken with the same rats after infection with trypanosomiasis, and experiments were made both with pathogenic and non-pathogenic trypanosomes. The pathogenic trypanosomes used were *T. equinum*, *T. brucei*, and *T. evansi*, and the non-pathogenic *T. lewisi*.

The following conclusions are drawn:—

"1. In white rats experimental trypanosomiasis cause no rise in body temperature.

"2. Pathogenic trypanosomes of high pathogenicity seem to decrease the power of thermal regulation in white rats under the influence of prolonged cold; but the non-pathogenic form, *T. lewisi*, does not show this effect.

"3. In white rats at ordinary temperature and under the influence of prolonged heat, none of the trypanosomes used in these experiments had any effect upon thermal regulation.

"4. From observations made upon the blood of infected rats it seems that conditions of prolonged heat and cold may possibly decrease the virulence of pathogenic trypanosomes without diminishing their pathogenicity."

(240) NOELLER (W.). **Die Uebertragungsweise der Rattentrypanosomen.**

II Teil. [The Method of Transmission of the Rat Trypanosome.

Part II.]—*Arch. f. Protist.* 1914. June 17. Vol. 34. No. 3. pp. 295-335. With 2 plates and 3 text-figures.

Conclusions:—

1. Fleas become infected more easily upon freshly infected rats than upon rats suffering from chronic trypanosomiasis.

2. The flea of the fowl can live for weeks upon rat blood. It does not come into consideration as an actual transmitter of the rat trypanosome, as it passes little or no faecal matter during the process of feeding.

3. The developmental forms of the rat trypanosome observed by PROWAZEK, BALDREY, and RODENWALT in the rat louse can be discovered with ease if the lice examined are taken from rats during the first week of infection.

4. In view of the fact that infectivity is soon lost after rat lice are removed from infected rats—according to GONDER in 20 days—the rat louse cannot be described as the true intermediate host of the parasite, if the view is held that a cycle of development must take place in the true carrier.

5. The rat louse excretes trypanosomes with the faecal matter.

6. Trypanosomes only find their way into the body cavity of the infected rat louse if the intestinal canal suffers injury.

7. Persistence of the trypanosomes in the rat louse, such as would be necessary for the production of a chronic infection of the louse, does not occur.

8. In the sheep ked (*Melophagus ovinus*), which can be fed upon rat blood for weeks, the trypanosomes retain their vitality for two days at 25° C., even if the ked has fed upon the blood of non-infected rats in the interval. Subsequently trypanosomes cannot be discovered in the ked. After 24 hours transition forms make their appearance in the ked, which resemble the so-called male trypanosomes from *Glossina*.

9. *Leptomonas ctenocephali* occurs in the larvæ of the dog flea also.

10. Among other parasites observed in the dog flea were: *Malpighiella refringens*, Minchin, and *Nosema pulicis*, Nöller. The gregarine *Actinocephalus parvus*, Wellmer, found in the dog flea larvae was probably derived from larvae of fowl fleas.

11. The following protozoa were observed in the fowl and pigeon fleas: *Leptomonas spec.*, which occurs very frequently in the hind gut of full-grown fleas and larvae; *Legerella parva*, Nöller, 1913, in the Malpighian vessels of an adult flea; *Actinocephalus parvus*, Wellmer, in the intestines of all the larvae examined.

- (241) BROWN (W. H.). **A Note on the Pathogenicity of *Trypanosoma lewisi*.**—*Jl. Exp. Med.* 1914. Apr. Vol. 19. No. 4. pp. 406-410.

Attention is drawn to the fact that other authors have placed upon record the occurrence of pathogenic and non-pathogenic strains of *T. lewisi*, both as regards the rat and as regards the mouse.

The author has encountered a strain of *T. lewisi* which for a time shewed a marked increase in virulence for the rat.

A strain of the trypanosome, the origin of which was not known, but which had been carried on for several years in white rats without shewing any unusual pathogenic effects, appeared to acquire a degree of virulence which rendered it fatal to rats by intraperitoneal inoculation with a drop or two of infected blood.

Of eight rats inoculated intraperitoneally with 0.2 cc. of defibrinated blood mixed with 0.8 cc. of salt solution, two died on the sixth day, one on the seventh, and the remainder on the ninth day after inoculation. The symptoms shewn were torpidity, weakness, anaemia, dyspnoea, in some cases blood-stained nasal and lachrymal discharges, and in all cases diarrhoea.

Careful examination failed to reveal any cause of death other than trypanosomiasis.

At the post-mortem examination the rats shewed acute enteritis, splenic enlargement, hyperplasia of the bone marrow, and a few necrotic foci in the liver. Some of the rats shewed congestion of the lungs, but in no case was there any pneumonia.

Immune and control rats remained healthy.

A number of observations have been made upon normal and immune rats inoculated with different strains of *T. lewisi*, and these shew that such conditions may occur frequently in a series of inoculations, while they do not occur in control or immune rats or in rats inoculated from other sources. With one exception symptoms of the same nature have not been observed in rats placed in contact with rats exhibiting them. In sub-inoculations the strain became less virulent again.

- (242) BROWN (W. H.). **Morphological and Developmental Anomalies of a Pathogenic strain of *Trypanosoma lewisi* and their Relationship to its Virulence.**—*Jl. Exp. Med.* 1914. June. Vol. 19. No. 6. pp. 562-569. With 2 plates comprising 22 figures.

The strain of *Trypanosoma lewisi* referred to in the previous paper was found to possess several morphological and developmental peculiarities. Attention was first drawn to the possibility of some connection existing between the pathogenicity and the morphology of *T. lewisi* by WENDELSTADT and FELLNER, who passed the trypanosome through cold-blooded animals and thus produced certain alterations in these characteristics. The author, therefore, followed up his pathogenic strain of *T. lewisi* through 82 rats, special attention being paid to the nature of the infection.

The details of experiments in which different series of infections were compared with parallel series in which a typical strain was used are reserved for another paper.

In the present paper only those deviations from the normal are described which were pronounced and occurred with sufficient frequency to be considered characteristic of the particular strain or infection. Most of the peculiarities have been observed before, but some of them are comparatively rare.

Two different types of multiplication forms were observed in addition to the usual forms. Figures are given which represent the two types, but it is a little difficult to appreciate what are the special characters of the multiplying forms. It is said that the trypanosomes may multiply very rapidly, producing a large number of irregular or small forms, or they may multiply slowly, the process continuing beyond the period at which it usually comes to an end.

Trypanosomes were found which possessed certain morphological features which were not normal. The variations observed involved all the structural features of the organism. The occurrence of trypanosomes containing no nuclei is referred to, and the occurrence of trypanosomes devoid of centrosomes is somewhat emphasised.

It is suggested that there is a correlation between the morphological and developmental characters and the virulence of the organism.

- (243) LAVERAN (A.) & ROUDSKY (D.). **Contribution à l'Étude de la Virulence du *Trypanosoma lewisi* et du *Tr. duttoni* pour Quelques Espèces Animales.** [The Virulence of *T. lewisi* and *T. duttoni* for Various Species of Animals.]—*Bull. Soc. Path. Exot.* 1914. June. Vol. 7. No. 6. pp. 528-535.

In this paper the authors give brief details of a number of experiments in which rodents of different species were inoculated with *T. lewisi* and *T. duttoni*, and the following summary is given:—

When small rodents such as wood-mice, voles, dormice, meriones, gerbils, jerboas, and guinea-pigs are inoculated intraperitoneally with the blood of rats rich in *T. lewisi*, there is as a rule produced an infection of variable intensity. In some cases the trypanosomes are very scantily present in the blood and disappear in 24 to 48 hours, and in other cases there is a well-marked infection, trypanosomes increasing in numbers, dividing forms being numerous, and the blood invasion being persistent. Even in those cases in which the infection is abortive, it is not a simple matter of the trypanosomes simply passing into the blood, but an actual infection, as the animals thereby acquire immunity to *T. lewisi*.

In rodents other than rats, inoculations in series with *T. lewisi* have not succeeded, but examination of the records of the infections of rodents with *T. lewisi* suggests that it would not be difficult to obtain strains of *T. lewisi* acclimatised for different species, in the same manner as one can obtain strains of *T. lewisi* inoculable in series for the mouse and strains of *T. duttoni* inoculable in series for the rat.

- (244) YORKE (W.) & BLACKLOCK (B.). **Observations on a so-called Cure for Trypanosomiasis.**—*Ann. Trop. Med. & Parasit.* 1914. Apr. 21. Vol. 8. No. 1. pp. 51-53.

At the request of the British South Africa Company the authors have tested a serum supplied by H. C. SIEG and reputed to be of great value for the treatment of trypanosomiasis. The conclusion drawn is that the sera supplied had not the slightest therapeutic value in animals experimentally infected with pathogenic trypanosomes.

- (245) YORKE (W.) & BLACKLOCK (B.). Antimony trioxide in the Treatment of experimental Trypanosomiasis.—*Ann. Trop. Med. & Parasit.* 1914. Apr. 21. Vol. 8. No. 1 pp. 55-72.

The antimony trioxide—"trixidin"—used in the experiments detailed in this paper was in part supplied by KOLLE and in part prepared by the authors.

The drug was administered by intramuscular injection in the majority of cases, but in some instances intraperitoneal and intravenous injections were made. For the intramuscular injections 10 and 40 per cent. suspensions in oil were prepared. For the intraperitoneal injections 1 per cent. suspensions in salt solution were employed. For the intravenous injections the drug in a very finely divided form was suspended in distilled water. This was allowed to stand for 18 hours, the liquid then being drawn off and estimated as to the quantity of the drug present in it.

The best results were obtained by intramuscular injection, but it was found that the amount of the drug absorbed was extremely small. Six months after administration it could be found at the seat of injection in an unaltered condition. The drug was immediately deposited from the suspension used for the intravenous injections and caused plugging of the veins.

A great disadvantage of the intramuscular administration was that in many cases it gave rise to the formation of sterile abscesses.

Small animals appear to be far more tolerant of the drug than large ones. At least 1 mgm. per 10 gm. body weight can be administered to rodents without untoward results. Such doses administered to donkeys and dogs frequently caused death, and far smaller doses were always followed by abscess formation at the seat of injection.

In cases in which the administration of the drug caused a disappearance of trypanosomes from the circulation there were sometimes relapses after very long periods, amounting to more than a hundred days.

The following strains of trypanosomes were used:—*T. rhodesiense*, *T. brucei*, *T. ugandae*, *T. evansi*, *T. equiperdum*, *T. congolense*, *T. equinum*, *T. gambiense*, and *T. lewisi*. With the exception of the last two of these all the strains appeared to be very susceptible to the drug, trypanosomes disappearing from the circulation within 48 hours after the injection. The strains of *T. gambiense* and *T. lewisi* proved to be very refractory. Relapses occurred even among the susceptible strains, and in the case of the following trypanosomes such relapses occurred after trypanosomes had disappeared from the blood for a period of 100 days or more: *T. rhodesiense*, *T. ugandae*, *T. vivax*, and *T. equiperdum*.

In agreement with what was observed regarding the curative effect of the drug upon *T. rhodesiense* and *T. gambiense*, it was found that it exerted a prophylactic effect against the former, but not against the latter.

- (246) LURZ (R.). Heilungsversuche mit Salvarsan bei Schlafkrankheit. [The Curative Treatment of Sleeping Sickness with Salvarsan.]—*Arch. f. Schiffs- u. Tropen.-Hyg.* 1914. May. Vol. 18. No. 10. pp. 335-339.

This paper records the results obtained in the treatment of 16

cases of the disease, most of which were in the advanced stages. The dose generally employed was 0.3 gm., and it was repeated in from one to four days, but on some occasions slightly larger doses were given.

In some cases repeated injections produced no symptoms, but in others there was fever, associated with pains in the head, thorax and abdomen, vomiting, and in one instance pains in the joints. The symptoms began to disappear in about five hours.

In every case the treatment caused a reduction in the size of the glands and a disappearance of trypanosomes from the blood.

The patients were kept under observation for periods varying from three and a half to five and a half months.

The following is a resumé of the results :—

One patient in whose spinal fluid trypanosomes had been found died. In six cases relapses occurred, but the general condition of the patients remained unaltered. In one case there was marked loss of condition, but trypanosomes were not found in either the blood or the cerebro-spinal fluid. In the remaining eight cases trypanosomes were not again observed.

Improved results were not obtained when mercury was used in conjunction with the salvarsan.

(247) **VORWERK. Versuche mit Salvarsan bei Schlafkrankheit.**

[Experiments with Salvarsan in the Treatment of Sleeping Sickness.]—*Arch. f. Schiffs- u. Tropen-Hyg.* May. Vol. 18. No. 10. pp. 329-335.

This paper gives the results obtained in the treatment of twelve cases of sleeping sickness in German East Africa. The majority (ten) of the patients shewed glandular swellings, but were otherwise in good condition (second stage). The remainder were at more advanced stages of the disease.

The drug was administered by intravenous injections in doses of 0.4 grammes.

The following conclusions are drawn :

1. A single administration of a moderate dose of salvarsan intravenously during the second stage of the disease causes the immediate disappearance of the trypanosomes from the glands and the circulation.

2. Relapses occur in almost every case after intervals varying from a fortnight to six months, whether single or repeated doses are administered.

3. The drug is contra-indicated in the case of patients in the third stage of the disease—that is to say, when they are in poor condition or show nervous symptoms, as it is dangerous.

4. Salvarsan is of less value for the permanent cure of the disease than atoxyl, treatment with which has not infrequently been followed by long periods during which the patients have been free from relapses.

(248) **MACFIE (J. W. Scott). On the Pathogenicity of the Trypanosome (*T. nigeriense*) from a Case of Sleeping Sickness from Eket, Southern Nigeria.**—*Ann. Trop. Med. & Parasit.* 1914. April. Vol. 8. No. 1. pp. 29-39.

This paper contains an account of further experiments with the trypanosome isolated by FORAN from cerebro-spinal fluid from a case of sleeping sickness at Eket.

The trypanosome was originally isolated by inoculation of a guinea-pig. Some of the experimental inoculations were made with the blood of two further guinea-pigs inoculated from this one, and also from a monkey apparently deriving its infection from the same source.

Of eight rats inoculated only one became infected, and in this animal only a single trypanosome was found on the 59th day.

Seven wild mice were inoculated. Four became infected, but the trypanosomes were very scantily present in the blood. The period of incubation was five to nine days. The disease ran a very chronic course, and did not appear to be responsible for death in any instance.

Three out of twelve guinea-pigs inoculated became infected. Parasites were fairly constantly present in the blood but never in very large numbers. The period of incubation was 27 days, and in one case the animal died on the 158th day.

In one dog out of seven trypanosomes were found on the 85th day and were subsequently found to be present in small numbers. The dog was alive and apparently in good health on the 115th day.

Two monkeys were successfully inoculated from the original guinea-pig. The infection was very chronic and the monkeys were still alive and apparently well on the 118th and 137th days.

One of two goats inoculated became infected. Trypanosomes were found to be very scantily present on two occasions only.

The evidence is said to be in favour of this trypanosome being a new species.

- (249) DELANOË (P.). *Des Variations du Pouvoir Infectieux et de la Virulence de Trypan. dimorphon* L. et M. (suite). Deuxième Note. [Variations in the Infectivity and Virulence of *Trypanosoma dimorphon* L. and M. Second Note.]—*Bull. Soc. Path. Exot.* 1914. Apr. Vol. 7. No. 4. pp. 281-289.

(For abstract of the Preliminary Note, see this *Bulletin*, Vol. 2, No. 1. p. 68.)

In this paper the author gives at length a description of the experiments carried out with a strain of *T. dimorphon* derived from a naturally infected ox, which at the outset was not inoculable for the white rat and the guinea-pig, and which after a series of experiments became pathogenic for both of these species and capable of causing a fatal infection in the former. The experiments do not as yet warrant any statement as to whether the trypanosome is fatal for the guinea-pig.

The strain acquired the power of infecting the rat after one passage through a kid and one through a dog. The strain became pathogenic for the rat after the first passage through the rat.

The author also found that when, after inoculation with the trypanosome, there was no blood invasion, the animal acquired no immunity.

The strain became infective for the guinea-pig after the following passages: one through a kid, followed by one through a dog, a second through a kid and one through a white rat.

The conclusion drawn is that it is difficult to demonstrate the susceptibility of the white rat and the guinea-pig to *T. dimorphon*, and that a trypanosome can be said to be non-pathogenic for a particular species only after repeated and varied experiments.

- (250) KLEINE (F. K.), FISCHER (W.) & ECKARD (B.). Ueber die Bedeutung der Speicheldrüseninfektion bei der Schlafkrankheitsfliege (*Glossina palpalis*). II Mitteilung. [The significance of the infection of the Salivary Glands in *Glossina palpalis*. Second Part.]—*Zeitschr. f. Hyg. u. Infektionskr.* 1914. May 27. Vol. 77. No. 3. pp. 495-500.

An abstract of the first communication on this subject will be found on page 282 of Volume I of this *Bulletin*.

In this paper the authors detail some experiments regarding the path by which trypanosomes (*T. gambiense*) reach the salivary glands.

The possibility was considered that it might be from the body cavity. Laboratory-bred flies were given four feeds upon monkeys infected either with *T. gambiense* or *T. rhodesiense* and were then fed upon healthy monkeys until these were found to be infected. The whole batch of flies used in the particular experiment were then killed by chloroform.

In order to obtain fluid from the body cavity two methods were adopted. In the majority of cases the legs were cut off close to the thorax, and the liquid made to exude from the abdominal and thoracic cavities by slight pressure upon the wounds with a slide. In the other cases a transverse slit was made about the middle of the under surface of the abdomen, and from this a cut was made upwards. The exuding liquid was collected in a capillary pipette, films being subsequently made from it.

In 21 experiments 215 flies were used, and of these at least 21 actually became infected.

A further batch of 207 flies which had had a single meal on an infected monkey and were then fed upon healthy animals were examined, but in this case the question of the infectivity of the flies was not actually determined.

In no cases were any trypanosomes found in the fluid obtained from the body cavities, although in the second batch of flies eleven were found to have numerous trypanosomes in their intestines, and were consequently on the way to becoming infective, if not actually so.

Previously to carrying out the experiments mentioned the authors had examined the fluid from the body cavities of nine infected flies and in three instances had discovered trypanosomes. They, however, believe that these discoveries were probably due to faulty technique.

- (251) HAGEMEISTER (Wolfgang). Ueber die Zuchtung pathogener Trypanosomen auf künstlichen Nährböden. [The Cultivation of Pathogenic Trypanosomes in Artificial Media.]—*Zeitschr. f. Hyg. u. Infektionskr.* 1914. Apr. 1. Vol. 77. No. 2. pp. 227-256.

The medium used by the author in his later experiments was as follows:—

Extract of 125 gm. of beef or horse flesh in 1000 cc. of distilled water.

20 gm. of agar.

20 gm. of peptone.

5 gm. of sodium chloride.

The reaction of the medium was neutralised and 5 cc. of normal soda solution added in excess. There was also added 2 per cent.

of dextrose on the lines recommended for the cultivation of malarial parasites and piroplasms.

To this medium two or three times the volume of sterile defibrinated blood was added. It was found that the addition of dextrose obviated the necessity of using rabbit blood, this being replaced successfully by goat, horse, donkey, and calf blood. The tubes were sealed with paraffin wax.

The trypanosomes used were the "Ferox" strain of nagana and the Eastern Prussian strain of *T. equiperdum*.

The seed material for the cultures consisted of heart blood of rats and mice which had been inoculated intraperitoneally. The blood was drawn from the still beating hearts of heavily infected animals into capillary tubes containing salt solution. Two or three drops of this mixture were used for the inoculation of each tube. The tubes were kept in the dark at room temperature.

Experiments were first made with the trypanosome of the *theileri* type occurring in cattle in Germany. It was found possible to obtain growths in those tubes only which contained ox blood and were incubated. Only 50 per cent. of these tubes shewed any growth, and in these there was active growth from the fifth day onwards. Trypanosomes began to decrease in numbers from the twelfth day and had disappeared by the eighteenth day. Sub-cultures were obtained in tubes of medium containing goat blood.

With the *T. brucei* the author was never able to find trypanosomes in his cultures after the eleventh day, and in no case were they numerous. There were no rosettes formed. A series of tubes to which goat blood was added succeeded in only 20 per cent. and in these trypanosomes were visible for four days only.

Parallel series of tests were made, and it was found that the tubes containing bloods other than rabbit blood and to which dextrose had been added yielded better results than the Novy medium as originally described.

The addition of dextrose to media containing rabbit blood did not improve them.

Conclusions :

Multiplication of ox trypanosomes of the *T. theileri* type on media containing ox blood and incubated at 37° C. occurs in the first generation only. But subcultivation succeeded with trypanosomes of this species when media containing goat blood were used.

The addition of dextrose has a beneficial effect upon the vitality of the trypanosomes and upon their capacity for multiplying in vitro.

The addition of dextrose to other kinds of blood (goat, horse, donkey, calf) not only replaced the expensive rabbit blood, but actually rendered them superior to it. Dextrose did not exert any beneficial effect upon media containing rabbit blood.

Trypanosomes cultivated upon media containing dextrose retained their infectivity, but lost their virulence to some extent.

A number of passages through cultures does not render the pathogenic trypanosomes more suitable for cultivation in vitro, but their vitality is thereby decreased and their virulence is rapidly diminished.

T. brucei retains its virulence in cultures better and with more regularity than *T. equiperdum*.

- (252) PRINGAULT (E.). *Cimex pipistrelli*, Jen., Agent de la Transmission de la Trypanosomiase des Chauves-souris. [*Cimex pipistrelli*, Jen., the Transmitting Agent of Trypanosomiasis in Bats.]—*Compt. Rend. Soc. Biol.* 1914. June 5. Vol. 76. No. 19. pp. 881-883.

The author has carried out experiments indicating that bugs act as the transmitting agent of *Trypanosoma vespertilionis*.

- (253) PRINGAULT (E.). Non-pathogenicité du *Trypanosoma vespertilionis* (Battaglia) pour les Animaux de Laboratoire. [The Non-Pathogenicity of *T. vespertilionis* for Laboratory Animals.]—*Compt. Rend. Soc. Biol.* 1914. June 5. Vol. 76. No. 19. pp. 883-884.

The author is in agreement with SERGENT, NICOLLE, and COMTE in finding this trypanosome non-pathogenic for laboratory animals, and disagrees with BATTAGLIA, who successfully infected guinea-pigs and rabbits.

- (254) MAYER (M.) & DA ROCHA-LIMA (H.). Zum Verhalten von *Schizotrypanum cruzi* in Warmblütern und Arthropoden. [*Schizotrypanum cruzi* in Warm-blooded Animals and Arthropods.]—*Arch. f. Schiffs- u. Trop.-Hyg.* 1914. Beiheft 5. Vol. 18. pp. 101-136. With 2 coloured plates.

Summary.

1. A description is given of the course of the infection when passed through monkeys, guinea-pigs and mice, and particular interest attaches to the occurrence of relapses after long periods (in one case after eight months in the guinea-pig).

2. In most cases recovery renders rats, rabbits, and guinea-pigs resistant to re-infection. The serum of such immune animals was found to have no therapeutic effect when injected 24 hours before and simultaneously with infective material.

3. Complement-fixation experiments with serum and extracts of organs from infected animals did not give any results different from those given by controls.

4. Dividing forms, save those already described by CHAGAS, in which nuclear division was just commencing, were never seen in the peripheral blood. Leishmanial forms were frequently observed.

5. Examination of sections shewed that the *Schizotrypanum* multiplies by division within the tissue cells, the parasites being rounded or spindle shaped as a result of pressure, and that the size and form of the parasite is retained during multiplication. Finally small individuals are produced at the last division which rapidly develop into trypanosomes. These leave the host cell after a short time (the time varying with the tissue) and invade the blood stream.

6. The normal development of the Schizotrypanum within the tissue cells does not appear to injure these in any way. Even the mechanical effects produced by large groups of parasites are rapidly and completely repaired, at least in muscle fibres. Toxic substances appear to be produced when the parasites are injured in any way. Altered host cells may contain parasites which have abnormal morphological and developmental features. There is no direct connection apparently between the formation of a colony of parasites in organs and symptoms of general degeneration or disturbance of function. A causal connection is only established when parasites are destroyed in large numbers. In such cases there may be destruction of the host cells and not infrequently evidence of wide-spread inflammatory changes. There is therefore no basis for explaining clinical symptoms and anatomical alterations on the grounds of the discovery of uninjured developing parasites.

7. Animals of the same kind, infected in the same manner, and shewing the same clinical picture, shew on microscopic examination of their organs the widest differences in the extent and severity of the lesions and in the distribution of the parasite. The most severe lesions are generally found in the stripped muscular tissue. The fatty tissue was frequently the seat of extensive degeneration. The other organs, although they frequently contained colonies of parasites, in most cases shewed no lesions.

8. The development of the parasite in *Conorhinus megistus*, *Acanthia lectularia* and *Ornithodoros moubata* was followed more closely. Transmission by biting was not determined with certainty in a large number of series (two doubtful positive results). The ingestion of infected ticks by mice and the bites of such ticks in no case caused infection.

9. No development occurred in *Stegomyia calopus* and *Culex pipiens*.

10. Experiments with the various "trypanotropic" drugs have yielded only unsatisfactory results up to the present.

(255) BLACKLOCK (B.). The Multiplication and Infectivity of *T. cruzi* in *Cimex lectularius*.—*Brit. Med. Jl.* 1914. Apr. 25. No. 2782. pp. 912-913

The experiments described were undertaken in order to ascertain whether *T. cruzi* is capable of existing and developing in *Cimex lectularius*, and if so, whether the forms found in the bed bug are capable of infecting laboratory animals when injected into them, or when the bugs feed on the animals.

Conclusions :—

"1. *T. cruzi* is capable of living and multiplying in *Cimex lectularius* for long periods.

"2. The parasites found in the bed bug are infective on inoculation as early as 21 hours and as late as 77 days from the infecting feed.

"3. It is not possible to say which of the many different forms occurring in the bug causes infection in the vertebrate host.

"4. Transmission of the disease to healthy animals by feeding infected bugs on them is of very rare occurrence. It was only once observed in the course of these experiments.

"5. There is no evidence of the hereditary transmission of *T. cruzi* in *Cimex lectularius*."

LEISHMANIASIS.

- (256) CARONIA & DI GIORGIO (G.). **Sulla Leishmaniosi spontanea nei Cani di Palermo.** [Spontaneous Canine Leishmaniasis in Palermo.] —*Pathologica*. 1914. Apr. 15. Vol. 6. No. 131. pp. 208-209.

This brief paper gives the results of the examinations carried out on dogs during the twelve months ending August, 1913. One thousand and five dogs were examined and only one was found to be infected.

- (257) PITTALUGA (G.). **Kala Azar Infantile e Leishmaniosi canina in Ispagna.** [Infantile Kala Azar and Canine Leishmaniasis in Spain.] —*Pathologica*. 1914. Mar. 1. Vol. 6. No. 128. pp. 121-123.

The author records the discovery of two cases of spontaneous canine leishmaniasis at Tortosa, and one at Beninar, in Almeria. Cultures were successfully made.

- (258) DA SILVA (P.). **Notes sur le Kala Azar.** [Notes on Kala Azar.] —*Arquiv. do Instit. Bact. Camara Pestana*. 1914. June. Vol. 4. No. 2. pp. 147-172. With 2 plates and 3 text figs.

During the period May, 1910, to March, 1911, 300 dogs were examined and eight were found to be infected with Leishmania. The spleen, bone marrow, and liver were examined. Subsequently seven other dogs were examined, and one found infected.

During the period March to June 1912, a fresh series of examinations was made and four infected dogs were found among 109 examined. In these instances only the spleen was examined.

Details are given of 12 dogs which were used for experimental inoculations, the virus in one experiment having been obtained in the first instance from a case of the disease occurring in a human being (a girl of nine years of age). In other cases the infection was transmitted by inoculation from a naturally infected dog to others.

In experimentally infected dogs no symptoms are said to be observed. A diagnosis must be based upon the detection of the parasite in the liver or in the bone marrow. It is difficult or impossible to locate the spleen by palpation and therefore spleen puncture was not resorted to. The author prefers the examination of bone marrow to that of material obtained by liver puncture as being more certain.

Cultivation upon NNN medium is more certain than microscopic examination for the detection of the parasite.

The belief is stated that the skin lesions that have been described as occurring in cases of canine leishmaniasis are due to follicular mange and not to the leishmaniasis.

It was found to be possible to transmit both the human and canine strains to dogs by inoculation.

In the author's opinion only two of the experimentally infected dogs actually died of the disease, death in other instances being due to follicular mange.

A number of experiments have been carried out regarding the transmission of leishmaniasis with fleas, both *Pulex irritans* and *Ctenocephalus canis*, and, though very definite results have not been obtained, the author appears to incline to the view that fleas play a part in the transmission.

- (259) PRINGAULT (E.). **La Leishmaniose canine à Marseille.** [Canine Leishmaniasis at Marseilles.]—*Bull. Soc. Path. Exot.* 1914. June. Vol. 7. No. 6. pp. 484-488.

During the period December to May the author has examined 310 dogs of various species and has found five infected with Leishmania. He gives his reasons for believing that these dogs have not been imported from places where canine leishmaniasis exists, and reaffirms his statement that canine leishmaniasis actually occurs at Marseilles.

- (260) NICOLLE (C.) & CONOR (Marthe). **Difficulté de Conservation du Virus de la Leishmaniose Canine par les Passages.** [The Difficulty of preserving the Virus of Canine Leishmaniasis by Passage.]—*Bull. Soc. Path. Exot.* 1914. June. Vol. 7. No. 6. pp. 481-484.

The authors referred to this difficulty in 1912 [see this *Bulletin*, Vol. 1, pp. 109-110], and the present paper contains a detailed description of a fresh series of experiments of the same nature as those originally described.

The following is a summary of the results obtained :—

At the first passage two dogs out of three were certainly infected, and three monkeys out of four were probably infected.

At the second passage one monkey out of four became infected, but the inoculation failed in the case of three dogs.

At the third passage two mice and one of two monkeys became infected.

At the fourth passage three monkeys and two mice were inoculated without success.

The canine virus appears to be more difficult to maintain by passage than the human virus, and it is a curious fact that the virus isolated from a naturally infected dog appears to be less virulent for the dog than the human strain.

The impossibility of protecting dogs from infection with piroplasmiasis in Tunis increases the difficulties of maintaining the virus.

- (261) NICOLLE (C.). **Chronique du Kala Azar en Tunisie.** [Kala Azar in Tunis.]—*Bull. Soc. Path. Exot.* 1914. June. Vol. 7. No. 6. pp. 479-481.

Since the publication of statistics in the Archives of the Pasteur Institute, Tunis, in 1912, eight additional cases of the disease have been observed in the human subject, making 38 cases in all. In addition, there has been one case recorded in a child in Algeria. All the cases save one occurred in children under three years of age, and six were in boys.

During 1913, 109 dogs were examined. The animals were principally selected from among animals whose appearance raised the suspicion that they might be infected. Six cases were detected, four severe and two moderate.

During September 20 cats were examined, but with negative results in every case.

Five strains of parasites are kept in artificial culture; two from cases of kala azar, two from naturally infected dogs, and one from a case of oriental sore. NNN medium is used, and subcultures are made twice monthly. The parasites have been maintained without morphological alteration through from 56 to 119 generations.

All the cultures grow easily and form little transparent colonies on the surface of the blood-agar.

The authors draw attention to the occurrence of splenic anaemia in children only a few months old, the cause of which has not been determined.

- (262) Row (R.). **Generalised Leishmaniasis induced in a Mouse with the Culture of *Leishmania tropica* of Oriental sore.**—*Bull. Soc. Path. Exot.* 1914. Apr. Vol. 7. No. 4. pp. 272-277.

The author has succeeded in producing a generalised infection by inoculating a mouse intraperitoneally with cultures of *L. tropica*. Three inoculations were given on three successive days, using cultures varying from four to seven weeks old, and a final inoculation was given two months later. The mouse was killed nine months after the final inoculation. The spleen was six to eight times the normal size, and the liver was distinctly enlarged. The peritoneum appeared to be normal. The parasites were present in large numbers in the spleen, liver, and bone marrow, and cultures were raised from them. Films of the heart blood showed "large cocci-like dots occupying apparently the red cells singly." That these bodies were not cocci was shown by the fact that they could not be cultivated upon agar.

This appears to furnish evidence that *L. tropica* and *L. donovani* may be identical, in spite of the different clinical aspects of oriental sore and kala azar.

- (263) WENYON (C. M.). **Demonstration by Dr C. M. Wenyon of some smears made by Dr. NELIGAN from Oriental Sore in Dogs in Teheran, Persia.**—*Trans. Soc. Trop. Med. & Hyg.* 1914. June. Vol. 7. No. 6. pp. 215-216.

"Dr. C. M. Wenyon showed some specimens, under the microscope, of smears made by Dr. NELIGAN from oriental sore in dogs, in Teheran, Persia, and made the following statement with regard to them:—

"Dr. NELIGAN, who was the first to demonstrate leishmania in the skin lesions of dogs, tells me that oriental sore is very common, both in human beings and in dogs, in Teheran. Dr. NELIGAN's discovery of oriental sore in dogs is of much interest in that it places this disease on the same footing as kala-azar, which is known to occur naturally both in human beings and dogs. A further observation which Dr. NELIGAN has made is that leishmania not only occur in the sores about the heads of the dogs, but that they also occur in the internal organs. Dr. NELIGAN has sent me smears of the organs of dogs which had oriental sore and in the bone marrow fair numbers of leishmania occur. Here is a difficult question to settle. Does the presence of leishmania in the organs mean that the dogs are suffering from a general infection with *Leishmania tropica*, the parasite of oriental sore, or does it mean that in Teheran there occurs also canine kala-azar, and that the dogs with the general infection, as well as the local skin lesions, are suffering from both diseases, oriental sore and kala-azar? A mere examination of the films cannot answer this question, which can only be settled on the spot by further observations and experimental work."

SPIROCHAETOSIS.

- (264) MACFIE (J. W. Scott) & JOHNSTON (J. E. L.). **A Note on the Occurrence of Spirochaetosis of Fowls in Southern Nigeria.**—*Ann. Trop. Med. & Parasitol.* 1914. Apr. Vol. 8. No. 1. pp. 41-45. With 2 plates.

The disease, which is well-known among the natives, was first brought to the notice of the authors as occurring in Muscovy ducks, but subsequently it spread to the fowls in the same compounds.

The symptoms were those usually observed in spirochaetosis. On post-mortem the liver and spleen were found to be greatly enlarged, the former containing necrotic patches of considerable size.

The parasite appeared to resemble very closely that described by BALFOUR as occurring in the Soudan.

Atoxyl and salvarsan were tried in a very small number of cases and the results appeared to indicate that salvarsan would yield the better results.

The authors were unable to convince themselves of the presence of "after phase" bodies described by BALFOUR, and had no opportunity to carry out any immunity tests with the object of determining the identity of the parasite.

- (265) LAUNOX (L.) & LÉVY-BRUHL (M.). **Le Sang de la Poule dans la Spirillose expérimentale.** [The Blood of the Fowl in Experimental Spirillosis].—*Ann. Inst. Past.* 1914. May. Vol. 28. No. 5. pp. 517-539. With one coloured plate.

Conclusions :—

1. Infection of adult fowls with *Spirochaeta gallinarum* sets up a very marked and rapid anaemia, the number of blood corpuscles being in some cases reduced to half the normal in five days. This phenomenon is the most characteristic from the haematological point of view. The intensity of the anaemia and some of the characters possessed by it cause spirillosis to resemble the so-called pernicious anaemias. But this disease differs from the pernicious anaemias in that there is a rapid and complete regeneration of the blood within a fortnight after the crisis.

2. The anaemia and the regeneration of the blood are accompanied by variations in the leucocytes which follow a constant cycle.

| Day of Disease. | Clinical. | Red Corpuscles. | White Corpuscles. |
|-----------------|---------------|-------------------------|--|
| 1 to 2 | Incubation .. | Commencing anaemia | Leucocytosis with polynuclears having rod-shaped granules. |
| 3 to 5 | During attack | Progressive destruction | Leucocytosis with neopolynuclears. |
| 5 to 6 | Crisis | Maximum anaemia .. | Leucopenia. |
| 6 to 10 | Convalescence | Regeneration. | Mononuclear leucocytosis. |
| „ | Recovery | Normal. | Normal, slight leucocytosis or sometimes prolonged mononuclear leucocytosis. |

3. The spleen appears to play no part in these modifications in the blood. Splenectomy does not influence the series of changes indicated in the table.

- (266) BRONFENBRENNER (J.). **A Simplified Method for Cultivating Spirochaetes on Liquid Media.**—*Proc. Soc. Exp. Biol. & Med.* 1914. June. Vol. 11. No. 6. pp. 185-187. With 1 text fig.

The method described is a modification of NOGUCHI's method, and was devised in order to obviate the use of a rubber stopper connection between the upper and lower parts of the apparatus, as this often becomes a source of contamination of the culture.

The new method may be used in two ways:—

1. A single tube replaces the two tubes joined by a rubber bung in NOGUCHI's method. A piece of rabbit kidney is placed in the bottom of the tube, which is then drawn out at about its middle point to form a constriction. It is then filled with ascitic broth or sheep serum water to the point where the tube broadens out again above the constriction. Another piece of tissue is placed in the upper portion of the tube and this is filled with ascitic agar, preferably 1 per cent. In this the spirochaete culture is placed. Sterile paraffin oil in a thin layer is then placed above the agar and the tube incubated.

2. A better way is as follows: A piece of tissue is placed in the bottom of a tube, which is then drawn out as before. By means of a capillary pipette the spirochaete culture and ascitic broth are introduced into the bottom of the tube. A bung is fitted into the mouth of the tube, through which passes a glass tube shaped like the letter Y, with the two upper arms projecting freely. One of these is connected to an exhaust pump and the other to a reservoir of sterile paraffin oil. The tube is exhausted by the pump, the tube being immersed in water at 37° C. to facilitate the exhaustion. After exhaustion, the cock below the oil reservoir is opened and the oil is allowed to run down on to the surface of the medium, the negative pressure in the tube assisting this. The tube is then sealed off at the constriction. It is said that the oil makes it possible to keep the spirochaetes alive after the tube is once opened. The author has kept his cultures going in this way for three months.

MALARIA AND MISCELLANEOUS HAEMO-PARASITES.

- (267) SEIDELIN (H.) & CONNALL (A.). **A Note upon the Occurrence of a Plasmodium in the Blood of West African Monkeys.**—*Ann. Trop. Med. & Parasit.* 1914. Apr. 21. Vol. 8. No. 1. pp. 81-82.

The authors have discovered parasites belonging to the genus *Plasmodium* in each of three young monkeys examined at the Medical Research Institute at Yaba, near Lagos. Two of the monkeys were *Papio sphinx*, and the other was a *Cercopithecus mona*. The parasites were very scantily present, but in two of them both gametocytes and schizonts were seen.

The following characters were observed: The pigment in fresh preparations was of a bright yellow colour, and in preparations stained with Giemsa it took on a brilliant green tint. The chromatin was looser in texture and stained more faintly than the chromatin in parasites of the human subject. The cytoplasm, and particularly that of the microgametocytes, stained very faintly.

Particular interest lies in the fact that only three monkeys were received and all were found to be infected.

- (268) SERGENT (Ed.) & BÉGUET (M.). *De l'Immunité dans le Paludisme des Oiseaux. Les pigeons guéris de l'Infection à *Haemoproteus columbae* ne sont pas immunisés contre elle.* [Immunity in Avian Malaria. Pigeons which have recovered from Infection with *Haemoproteus columbae* have not acquired Immunity against it.] —*Compt. Rend. Soc. Biol.* 1914. June 12. Vol. 77. No. 20. pp. 21-23.

The authors find that in those rare cases in which recovery takes place rapidly no degree of immunity is acquired; but in those more usual cases in which recovery is not complete, parasites remaining in the blood for long periods, this latent infection corresponds to a condition of relative immunity. In the event of complete recovery taking place this relative immunity is lost.

- (269) ROWLEY-LAWSON (Mary). *Free Malarial Parasites and the Effect of the Migration of the Parasites of Tertian Malarial Infections.* — *Jl. Exp. Med.* 1914. June. Vol. 19. No. 6. pp. 523-535. With 6 plates (one in colour).

"1. The malarial parasite is extracellular throughout its life-cycle and migrates from red corpuscle to red corpuscle destroying each before it abandons it; in the brief intervals between, the parasite is free in the blood serum; it does not remain long free, but almost immediately attaches itself to another red corpuscle by means of delicate pseudopodia.

"2. The compact form of the tertian parasite is the type of free parasite most often observed; in this form the parasite may be seen not only in migration, but after quinine and in the cadaver. I believe the compact form to be the normal resting form of the parasite, all other forms being assumed in order to secure attachment and to obtain food.

"3. Care must be taken not to confound free parasites having protoplasmic pseudopodia ready for attachment with the sexual flagellating parasites, whose flagella are composed of chromatin.

"4. The malarial parasite can live for some time free in the blood serum, though under normal conditions there is no reason why it should remain free for any length of time, and there are certain periods in the life of the parasite when it must be admitted that it is free from the corpuscle and survives. If the parasite is, as I believe, attached to the external surface of the red corpuscle, it is constantly exposed to the action of the patient's serum.

"5. The destruction of more than one red corpuscle by each parasite would readily account for the severe and early anaemia occurring in malarial infections."

"6. Long continued treatment with quinine will eventually cause the death of all malarial parasites."

- (270) ROWLEY-LAWSON (Mary). **A Stage in the Migration of the Adult Tertian Malarial Parasite. Evidence of the Extracellular Relation of the Parasite to the Red Corpuscle.**—*Jl. Exp. Med.* 1914. May. Vol. 19. No. 5. pp. 450-458. With 5 plates (one in colour.)

The author recapitulates her views regarding the extra-cellular position of the malarial parasite and its migrations from corpuscle to corpuscle, and in the present paper refers particularly to that stage in which pigmented parasites are found partly on and partly off degenerated red corpuscles, caught apparently in the act of leaving them.

"1. What appear to be certain definite stages in the migration of the malarial parasite from red corpuscle to red corpuscle may be demonstrated by thorough and persistent observations, not minutes spent on each specimen, but many hours.

"2. The migration of the malarial parasite from red corpuscle to red corpuscle gives a reasonable explanation of the loss of red corpuscles which cannot be accounted for by the destruction of the infected corpuscles at the time the parasites segment.

"3. Migration to other red corpuscles is a satisfactory explanation of the ultimate fate of the young parasites seen in instances of multiple infection of single corpuscles.

"4. In the light of the facts here presented, it would seem impossible to explain the instances of the parasites partly on and partly off degenerated red corpuscles as the result of technique. A stage in the migration of the parasite seems to me to be the probable interpretation of the phenomenon.

"5. A corpuscular mound encircled by an adult parasite, when seen at the periphery of the red corpuscle, should have the same significance and interpretation, in reference to the extracellular relation of the parasite to the corpuscle, that it has when it is seen in connection with a young parasite.

"6. The adult ring-form parasite should have the same interpretation as the young ring-form parasite.

"7. Attachment to the external surface of the red corpuscles seems to me to be the only possible interpretation of the appearances of the parasites pictured in this article."

- (271) FLETCHER (W.). **The Wassermann Reaction in Malaria.**—*Lancet.* 1914. June 13. pp. 1677-1678.

Conclusion: The presence of malarial parasites in the blood does not cause the serum to give a positive Wassermann reaction when the method of BROWNING, CRUIKSHANK, and MCKENZIE is employed.

- (272) NAWROTZKY (N. N.). **Haematoparasitologische Notizen. Zweite Mitteilung.** [Notes on Blood Parasites. Second Part.]—*Centralbl. f. Bakt.* 1 Abt. Orig. 1914. Apr. 25. Vol. 73. No. 6. pp. 358-362. With 1 plate comprising 14 figures in colour.

This paper deals with the following parasites: The trypanosome of the pike, the trypanoplasma of the pike, a haemogregarine of the pike (new species), haemogregarine of the sterlet (new species), filaria of the white heron (new species), and the microfilaria of the rook.

- (273) RINGENBACH (J.). **Quelques Hematozoaires du Congo.** [Some Haematozoa from the Congo.]—*Bull. Soc. Path. Exot.* 1914. Apr. Vol. 7. No. 4. pp. 296-299.

The author has discovered a trypanosome in the blood of the
(669) o

hornbill, *Bycanistes albotibialis*, commonly known as the Calao, which is possibly different from the parasites described by a number of other authors. But, as the authors point out, morphological differences are not always sufficient to establish a species.

The trypanosome had the following average measurements :—

| | | | | | | |
|--|----|----|----|----|----|-------------|
| Total length | .. | .. | .. | .. | .. | 53 microns. |
| Maximum width | .. | .. | .. | .. | .. | 8 „ |
| Distance from the centrosome to the posterior extremity | .. | .. | .. | .. | .. | 5·7 „ |
| Distance from the centrosome to the anterior edge of the nucleus | .. | .. | .. | .. | .. | 14·3 „ |
| Larger diameter of the nucleus | .. | .. | .. | .. | .. | 4·2 „ |
| Free portion of the flagellum | .. | .. | .. | .. | .. | 8 „ |

A microfilaria has been found in the blood of the same bird. The parasite was 234 microns in length by 3 in width. In the fresh state it was very actively motile. There was no sheath. The body tapered towards the posterior extremity, which was bluntly rounded. The parasite appears to be distinct from those described by LEGER as occurring in various hornbills in the Soudan. The author proposes the name *Microfilaria bycanisti*.

A microfilaria has also been found in the blood of the Touraco (*Schizorhis cristata*). In the fresh state this parasite moves very slowly. It is provided with an envelope which scarcely exceeds the parasite in length. The body measures 127 microns in length by 3·7 in width; the sheath is about three times the width (10 microns). The name *Microfilaria schizorhinos* is suggested, as the author is unable to say whether the parasite is identical with that which PLIMMER found, but did not describe, in two touracos in West Africa.

The author has found parasites resembling the microgametocytes of *Plasmodium kochi* in the blood of a *Cercopithecus cephus*.

TSETSE FLIES.

(274) LLOYD (Ll.). Note on Scratching Birds and Tsetse-fly.—*Ann. Trop. Med. & Parasit.* 1914. Apr. 21. Vol. 8. No. 1. p. 83.

The suggestion has been made from time to time that guinea-fowl might, among other birds, act as a control to *Glossina* by ingesting the pupae, and the value of this suggestion has been tested by the author by the examination of the contents of the crops of ten guinea-fowl at Nawalia in the Luangwa Valley.

The birds are very numerous in Northern Rhodesia and especially in the Luangwa Valley.

In only three were any insects found, and in these they were very scantily present. No pupae of any kind were recorded. It appears that the bird is a vegetable feeder, and only eats insects when searching for food. The pupae of *G. morsitans* are usually placed in positions that are inaccessible to these birds, and therefore the guinea-fowl cannot be considered as suitable to act as a control of the fly.

- (275) SHIRCORE (J. C.). **Suggestions for the Limitation and Destruction of *Glossina morsitans*.**—*Bull. Entom. Res.* 1914. Apr. Vol 5. No. 1. pp. 87-90. With 1 map.

The author's observation leads him to conclude that in the "proclaimed area" there are at least four "primary centres" which harbour tsetse flies.

These areas are defined as "primary centres" for the following reasons.—Flies are found abundantly in these centres throughout the year, even when there are none or very few elsewhere. These are the only areas in which during the dry season there is water above or at a slight depth below the surface of the earth. These situations offer the best chance of feeding for the flies during the drought, as they are frequented by various species of game. The conditions, viz., light forest with open glades, short grass and water close at hand, form an ideal combination for both game and fly.

The probability is that the flies breed in these areas and from them spread to the surrounding country when the conditions become favourable. This spreading has been observed, and it has also been observed that "secondary centres" are formed with the onset of the rains, a different type of country being then more suitable.

The author advises that the forest connections along which these radiations take place should be cut off or otherwise dealt with about May, so that the "primary centres," which should have been delimited at the height of the previous dry season, may be isolated. The trees in the areas should then be cut down and left to dry, extensive burning of the areas being practised when the drying is complete.

- (276) ZUPITZA (M.). **Versuche und Vorschläge zur Verbesserung von Glossinenfangmethoden.** [Experiments and Suggestions for the Improvement of Methods for Catching Tsetse Flies.]—*Arch. f. Schiffs. u. Trop.-Hyg.* 1914. June. Vol. 18. No. 11. pp. 363-375. With 2 figures.

The author is of the opinion that the lined cloths used by CLEVE fail in their object to a great extent because they do not allow the passage through them of the scent of the man or animal upon which they are placed. To avoid this objection he used sheets of wire gauze with a mesh of about 2 centimetres. He found, however, that this was too large a mesh, and therefore used two layers of the gauze, one over the other. The gauze was hung in front and behind after the manner of sandwich boards, and the straps were so constructed that the gauze could be taken off for the purpose of removing flies adherent to it.

The lime used smelt of resin, and had a honey-like consistence and colour. During the warm periods of the day it became very fluid and had to be renewed every two hours. It was easily soluble in spirit, but not in pure water.

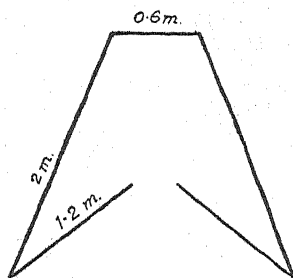
A comparison is made between the number of flies caught by means of this apparatus and the number caught during the same periods and at the same places with a net. This indicates that the butterfly net is slightly superior as a means of catching the flies.

Improvements are suggested in the manner of carrying the wire, mesh coverings. In order to completely cover the body it is suggested that a slightly flattened cylinder of the gauze would be the best. The gauze should be made of iron wire about 2 mm. thick, and should have a mesh of about 1 centimetre. Porous material of a rather dark colour should be worn underneath the gauze, and specially stout material should be used for covering the arms and legs.

The author also describes a fly trap with which no lime is used.

The trap consists of a cage, which in section is described as being shaped like an arrow-head with the point cut off. The depth of the trap from before to behind is about 2 metres. The side opposite that representing the truncated point of the arrow is composed of two doors opening inwards and leaving between them a narrow opening. The flycatcher is placed inside the trap. Flies coming in contact with the sides of the trap eventually find their way in and are caught. The trap itself consists of a framework covered with a thick muslin.

The diagram shows the plan of the trap with measurements.



- (277) SIMPSON (J. J.). **Entomological Research in British West Africa.**
V. Gold Coast.—*Bull. Entom. Res.* 1914. Apr. Vol. 5. No. 1.
 pp. 1-36. With 4 plates and 1 map.

This paper deals mainly with the distribution of various blood-sucking insects, but information is also given regarding the following points:—The position, configuration, and vegetation of the colony, climate and rainfall, insect-borne diseases in man and other animals, distribution of the genus *Glossina*, and parasites of mammals.

With regard to the diseases of animals, it is said that very few cattle are bred in the colony, but that 80 per cent. of the animals brought down from the north for slaughter at Coomassie are infected with trypanosomes. Only *T. cazalboui* and *T. pecaudi* have been found in cattle. BEAL states that these two with *T. dimorphon* are equally prevalent in horses. *T. dimorphon* alone has been found in dogs. Two sheep out of 83 examined have been found infected with trypanosomes similar to those occurring in cattle. Only one case of trypanosomiasis has been observed in the goat, and none in the pig.

Two cases of spirochaetosis have been recorded in the sheep and one in the goat by O'BRIEN.

Blood smears from eleven different kinds of game were examined for parasites, but none were found.

HELMINTHS.

- (278) RAILLIET (A.), HENRY (A.) & BAUCHE (J.). *Sur les Amphistomiens des Ruminants domestiques de l'Annam.* [The Amphistomes of Domestic Ruminants in Annam.]—*Recueil Med. Vet.* 1914. May 30. Vol. 91. No. 10. pp. 195-201.

In cattle slaughtered at Hue amphistomes are found most frequently in the rumen, but they also occur in the reticulum, abomasum, and bile ducts. A number of different species have been found.

In the buffalo the following parasites have been found in the rumen : *Gastrothylax cobboldi*, *G. elongatus*, *G. minutus*, *Paramphistomum orthocoelium*, and *P. papillosum*. In the ox, *Gastrothylax crumenifer* has been found.

G. elongatus has been found in the reticulum of the buffalo only.

Paramphistomum bathycotyle, *Cotylophoron cotylophorum*, and *Homalogaster paloniae* have been found in the abomasum of the buffalo, ox and sheep respectively.

Paramphistomum explanatum has been found in the bile ducts of the buffalo.

The authors think that *Paramphistomum fraternum*, Stiles and Goldberger, 1910, is synonymous with *P. explanatum*.

They are further of the opinion that *H. poirieri*, Giard and Billet, 1892, and *H. philippinensis*, Stiles and Goldberger, 1908, are in reality identical with *H. paloniae*.

The question of the pathogenic rôle played by amphistomes is considered, and the authors state that by means of the phenolphthalein test they have been able to detect the presence of blood in the bodies of *G. elongatus* and *crumenifer*, and have obtained doubtful results with *G. cobboldi* and *minutus*. Some evidence has been obtained that the ventral pouch of the parasites contains sanguinolent liquid. The question as to how this liquid gains access to the pouch is interesting, because it is known that neither the mouth nor the orifice of the ventral pouch is provided with an armature for penetrating the mucous membrane, nor has any lesion of the mucous membrane ever been described. The suggestion is made that the examination of parasites *in situ* in animals freshly slaughtered also throws some light upon the question of the nutrition of the parasites.

- (279) SEURAT (L. G.). *Sur un Nématode parasite du Flammant rose.* [A Nematode Parasite of the red Flamingo.]—*Compt. Rend. Soc. Biol.* 1914. May 22. Vol. 76. No. 17. pp. 814-817. With 4 text figs.

Fifteen specimens of a female *Tropidocerca* were found by the author in the gastric glands of two flamingoes killed in Algeria. Owing to the bright red colour, the name *Tropidocerca coccinea* is suggested.

Female.—Practically the whole of the body consists of a red globular structure measuring about 2.5 millimetres in diameter. Laterally, dorsally, and ventrally there are longitudinal bands of muscle fibres. These longitudinal bands are connected by circular strands which produce an appearance of transverse striation.

This globular portion of the body in reality represents only the middle part, greatly distended owing to the exaggerated development of the intestine and uterus. Anteriorly the body is continued in the

form of a pointed projection comprising the oesophageal and cephalic portions. Posteriorly there is a conical tail, and the anus is situated a little in front of the extremity.

The buccal cavity is rounded and measures about 20 microns. The oesophagus is encircled at its middle point by the nerve ring. The muscular portion of the oesophagus measures about 350 microns, and the glandular portion about 1.5 mm. The cephalic portion shews two lateral papillae anterior to the nerve ring; these are not exactly opposite each other. The ventral excretory pore opens very close behind the posterior border of the nerve ring.

The vulva is situated about 0.5 mm. in front of the anus. The vestibule and sphincter of the ovijector are very short. The vestibule is thickened in the distal portion, and contains nucleated muscle cells. It is lined by tall epithelial cells, which come into contact with each other at the free surface, leaving no visible lumen.

The uteri are very narrow, and are lined with large flat epithelial cells. They run parallel with each other and follow a very sinuous course. Numerous eggs are contained in them and the initial part of each is connected with the oviducts by a seminal receptacle of enormous size. The oviduct which leads from it is very narrow. The oviducts and the ovaries may be as much as 8 mm. in length.

The eggs have a thick smooth envelope, and the larvae at the time of hatching measure about 30 microns by 15. They collect in the uterus and pass through the ovijector without stopping there. About 50 may be found in each of the paired branches of the uterus.

The male is unknown.

MISCELLANEOUS.

- (280) WATSON (E. A.). **Preliminary Studies and Experiments with Swamp Fever.**—*Dept. of Agriculture, Canada. Report of the Veterinary Director General (F. Torrance, B.A., D.V.S.) for the Year ending 31st Mar., 1913.* Ottawa: Printed by C. H. Parmalee. pp. 86-87.

The author's experience regarding the virulence of swamp fever blood when inoculated in series in horses agrees with that of Van Es and others in that there is a progressive loss. A sample of blood obtained from a natural case in the Onoway District, Alberta, produced an acute attack at the first and second passages, but subsequently only chronic forms of the disease, which terminated in recovery. In these animals clinical symptoms were practically absent.

It is suggested that this is an indication that under natural circumstances the disease is insect borne, the insect cycle being necessary for the retention of the full degree of virulence.

It has been found that swamp fever blood does not react to the serum tests used for trypanosome diseases, but that the globulin content of the blood is markedly increased.

- (281) PANISSET (W. L.). **Paralysie bulbaire Infectieuse. Pseudo-Rage, Maladie d'Aujeszký.** (Infectious Bulbar Paralysis. False Rabies, Aujeszký's disease.)—*Rev. Gen. Med. Vet.* 1914. June 1. Vol. 23. No. 275. pp. 601-618. With 4 text figs.

This article contains no new information regarding the disease, but summarises our knowledge of the condition.

- (282) JONESCO-MICHAÏESTI (C.) & COMBIESCO (D.). **Sur une Epidémie de Dysenterie Bacillaire chez des Singes inférieurs.** [An Epidemic of Bacillary Dysentery in the Lower Monkeys.]—*Compt. Rend. Soc. Biol.* 1914. May 22. Vol. 76. No. 17. pp. 827-829.

The outbreak described by the authors occurred among a number of *Macacus rhesus* kept for experimental purposes. The clinical features and the lesions presented no remarkable features.

The organism isolated on Conradi-Drigalski and in bile broth resembled the Flexner type of bacillus.

Only two sera were available for agglutination tests: a polyvalent serum prepared with the bacilli of Shiga, Flexner, Dopter, Duval, and Y, and a serum agglutinating only the Shiga type. The Shiga serum agglutinated the bacillus isolated in dilutions of 1 in 80 only. The polyvalent serum agglutinated the Flexner and Y types in dilutions of 1 in 300, and the Strong type and the bacillus isolated in 1 in 150.

The authors conclude that:

With regard to the majority of the cultural characters, the bacillus isolated belongs to a type of dysentery bacillus allied to Flexner's bacillus, while as regards agglutination it approaches more closely the bacillus of Strong.

- (283) LANDOIS (F.) & HOEPKE (H.). **Eine endoparasitäre Milbe in der Lunge von *Macacus rhesus*.** [An Endoparasitic Acarus in the Lungs of *Macacus rhesus*.]—*Centralbl. f. Bakt.* 1. Abt. Orig. 1914. Apr. 25. Vol. 73. No. 6. pp. 384-395. With 1 plate and 3 text figs.

After summarising the recorded cases of endoparasitic acariasis the authors describe one occurring in the monkey.

The animal in which the lesions were found was one that was being used in a bone-transplantation operation at Breslau.

In each lung there were found eight to ten round nodules lying close under the pleura. They were not connected with a bronchus. In colour they varied from greyish-yellow to pale yellow, and on section shewed small, dry-looking centres about the size of lentils. The larger ones had crumbling material in the centre in which the acari could be detected with the microscope.

Histological examination shewed that the smallest nodules were composed of rounded and spindle-shaped cells, with a few leucocytes. At places there were collections of amorphous pigment granules of a reddish-brown colour. Tests of this pigment with potassium ferrocyanide gave negative results. The acari were found in the granular centres of the larger nodules, but none was found in the small nodules.

Material containing a single acarus was injected into the thoracic cavity of another monkey. Six months later this animal died and a single acarus was found in a nodule at the hilus of the left lung.

A detailed description of the parasite is given and the name *Pneumotuber macaci* is suggested for it.

- (284) BOUQUET (A.) & NÈGRE (L.). *Sur la Nature mycosique du Parasite de la Lymphangite epizootique. Formation de Gouttelettes d'Huile et de Filaments.* [The Mycotic Nature of the Parasite of Epizootic Lymphangitis. The Formation of Droplets of Oil and of Filaments.]—*Bull. Soc. Path. Exot.* 1914. June. Vol. 7. No. 6. pp. 464-466. With 10 text figs.

The authors bring forward the following evidence in support of their view regarding the blastomycotic nature of the *Cryptococcus* of RIVOLTA :—

In specimens prepared from tubes of agar medium which have been thickly sown with pus, incubated at 24°C. for a week, and then kept at room temperature for two months, the following vegetative forms have been observed :

In addition to the ordinary budding forms of the parasite there are certain organisms which have attained a size of twice the normal. These are found to contain refractile droplets or fine granulations. At the narrower pole of such parasites there is a cylindrical outgrowth which is granular and measures 2 microns in diameter. The longest outgrowth seen measured 18 microns and was divided by a septum into two equal parts. Similar large forms without any outgrowths may be found in the water of condensation and in milk inoculated with pus.

The refractile granules are spherical and at first small, but they may run together and form a large droplet nearly filling the *cryptococcus*. The oily nature of these droplets has been ascertained by staining with osmic acid.

- (285) BALFOUR (A.). *The Wild Monkey as a Reservoir for the Virus of Yellow Fever.*—*Lancet.* 1914. Apr. 25. pp. 1176-1178.

Attention was directed to this subject on account of a negro belief in Trinidad that an outbreak of yellow fever can be foretold because large numbers of red howler monkeys are always found dead before such an outbreak occurs, and certain information obtained suggests that there is possibly some connection between the deaths among the monkeys and the occurrence of yellow fever in man. It is suggested that workers in other parts of the world should investigate the subject.

(In the correspondence columns of the *Lancet* for May 9, 1914, Low writes to the effect that monkeys can but rarely if ever be reservoirs for the yellow fever virus, and brings forward evidence of his own to support this view.)

- (286) GIEMSA (G.). **Zur Schnellfärbung (Romanowsky-Färbung) von Trockenausstrichen.** [The Rapid Staining (Romanowsky Staining) of Dried Films.]—*Centralbl. f. Bakt.* 1. Abt. Orig. 1914. May. Vol. 73. No. 7. pp. 493-496. With 1 text fig.

In order to avoid certain defects in the method of rapid Giemsa staining published in 1910 the author has devised a new mixture which has the following composition:—

| | | | | | |
|----------------|----|----|----|----|-------|
| Azur II.-Eosin | .. | .. | .. | .. | 3 gm. |
| Azur II. | .. | .. | .. | .. | 0.3 „ |
| Glycerine | .. | .. | .. | .. | 25 „ |
| Methyl alcohol | .. | .. | .. | .. | 475 „ |

The solution is prepared warm, allowed to cool, and, after standing, is filtered. The solution is supplied under the name "*Farbfixerlösung nach G.*"

Slides having very thin films upon them are laid with the film side uppermost in special staining troughs which have ridges running across the bottom for the support of the slides.

The fixing and staining solution is dropped on to the slide from a drop-bottle until the whole of the film is covered, care being taken that the slide is absolutely horizontal. The trough is covered with a lid and left for half a minute, and in no case more than one minute.

In a wide measuring cylinder a mixture of 10 cc. of distilled water and 10 drops of the staining and fixing solution (*Farbfixerlösung*), or 10 drops of the original Giemsa solution, is prepared and this is poured into the trough so as to completely cover the slide. Gentle rocking completes the mixing of the solutions. The stain is then allowed to act for 10 minutes.

The specimen is washed in water, mopped, dried and mounted in liquid paraffin or in balsam that is free from acid.

This method differs from previous ones not only in the composition of this stain, but in the use of a staining mixture instead of water after the primary action of the staining and fixing fluid is completed. The object of this is to avoid the extraction of the stain from the specimen which occurs when water is used.

In the case of thick preparations they should be placed in distilled water or salt solution for a minute and again dried before staining, because this tends to dissolve the serous portion of the film which otherwise may become stained. Spirochaetes are said to be rendered especially distinct by this method.

The method is not so good for moist fixed preparations and for sections as the old method.

RECENT LITERATURE.

[Continued from this *Bulletin* Vol. 2, No. 2, pp. 110-112.]

Babesiasis (Piroplasmosis).

- (287) MISSON (L.). Immunisation Artificielle contre la Piroplasmose du Bétail Européen importé au Brésil. [The Artificial Immunisation of Cattle imported into Brazil from Europe against Piroplasmosis.]—*Folia Microbiologica*, 1914. June 27. Vol. 3. No. 1, pp. 49-75. With 3 plates.

Foot and Mouth Disease.

- (288) FAVERO (F.). Su la presenza di una sensibilizzatrice antiaftosa. [The Presence of a Sensitiser in Foot and Mouth Disease.]—*La Clinica Veterinaria*, 1914. Apr. 15-30. Vol. 37. No. 7-8, pp. 327-331.
- (289) KALLERT (E.). Untersuchungen über Maul- und Klauenseuche. I. Mitteilung. Ueber die Bedeutung der v. Betegh-schen Körperchen in der Aphthenlymphe. [Investigations into Foot and Mouth Disease. 1st Part. Significance of the von Betegh Bodies in the Lymph of the Aphthae.]—*Arbeit. a. d. Kaiserl. Gesundheitsamte*, 1914. June. Vol. 47. No. 4, pp. 591-602. With 2 plates.
- (290) KALLERT (E.). Untersuchungen über Maul- und Klauenseuche. II. Mitteilung. Beiträge zur Histogenese und Histologie der Maul- und Klauenseucheblase, insbesondere auch zur Frage des Vorkommens von Einschlusskörperchen in den spezifisch veränderten Teilen bei Maul- und Klauenseuche.—[Investigations into Foot and Mouth Disease. 2nd Part. Histogenesis and Histology of the vesicles in Foot and Mouth Disease, especially in regard to the Occurrence of Inclusion Bodies in the specifically changed Parts.]—*Arbeit. a. d. Kaiserl. Gesundheitsamte*, 1914. June. Vol. 47. No. 4, pp. 603-613. With 2 plates.
- (291) WEHRLE (E.) & KALLERT (E.). Versuche mit "Trypasafrol" und "Novotrypasafrol" sowie mit "Ernanin" bei Maul- und Klauenseuche. [Experiments with "Trypasafrol," "Novotrypasafrol," and "Ernanin" in Foot and Mouth Disease.]—*Berl. Tierärzt. Wochenschr.*, 1914. April 9. Vol. 30. No. 15, pp. 253-257.

Leishmaniasis.

- (292) CARINI (A.). L'Émétique dans le Traitement de la Leishmaniose cutanée et muqueuse. [Emetic in the Treatment of Leishmaniasis of the Skin and Mucous Membranes.]—*Bull. Soc. Path. Exot.*, 1914. April. Vol. 7. No. 4, pp. 277-281.

Malaria.

- (293) PITSCHUGIN (P. I.). Kultivierungsversuche mit *Plasmodium vivax* nach der Methode von Bass. [Cultivation Experiments with *Plasmodium vivax* according to Bass's Method.]—*Centralbl. f. Bakt.*, 1. Abt. Orig., 1914. Apr. 25. Vol. 73. No. 6, pp. 373-384.
- (294) STEPHENS (J. W. W.). A New Malaria Parasite of Man.—*Proc. Roy. Soc.*, 1914. April 8. Vol. B. 87. No. B 596, pp. 375-377. With 3 plates.
- (295) THOMSON (D.). The Origin and Development of Gametes (Crescents) in Malignant Tertian Malaria; some Observations on Flagellation, etc. (Being Part II of the Reports on the Malarial Expedition to Panama, 1912.)—*Ann. Trop. Med. and Parasit.*, 1914. Apr. 21. Vol. 8. No. 1, pp. 85-104. With one coloured plate.

Rabies, False.

- (296) SANGIORGI (Giuseppe). La Filtrabilità del Virus della Pseudorabbia.—*Giorn. R. Accad. Med. di Torino*, 1914. Mar.-Apr. Vol. 77. No. 3-4, pp. 69-78.
- (297) SANGIORGI (Giuseppe). Ricerche sulla Pseudorabbia.—*Pathologica*, 1914. June 1. Vol. 6. No. 134, pp. 282-284.

Trypanosomiasis.

- (298) CARINI (A.) & BOTELHO, jnr. (C.). Sur quelques Trypanosomes d'Oiseaux du Brésil. [Some Bird Trypanosomes of Brazil.]—*Bull. Soc. Path. Exot.*, 1914. May. Vol. 7. No. 5, pp. 395-398. With 3 text figures.
- (299) CARINI (A.) & MACIEL (J.). Existence de la Maladie de Chagas dans l'Etat de São Paulo. [The Existence of Chagas disease in Sao Paulo (Brazil).]—*Bull. Soc. Path. Exot.*, 1914. Apr. Vol. 7. No. 4, pp. 289-292.
- (300) LEGER (A.) & LEGER (M.). Trypanosomes et Haemoproteus d'Oiseaux du Haut-Sénégal et Niger. [Trypanosomes and Haemoproteus of Birds of Upper Senegal and Niger.]—*Bull. Soc. Path. Exot.*, 1914. June. Vol. 7. No. 6, pp. 493-497. With two tables.

Biting Flies.

- (301) AUSTEN (E. E.). A Dipterous Parasite of *Glossina morsitans*.—*Bull. Entomol. Research*, 1914. Apr. Vol. 5. No. 1, pp. 91-93. With 1 text figure.
- (302) FROGGATT (W. W.). Sheep-Maggot Flies in Australia. *Bull. Entomol. Research*, 1914. Apr. Vol. 5. No. 1, pp. 37-39.
- (303) LLOYD (Ll.). Further notes on the Bionomics of *Glossina morsitans* in Northern Rhodesia.—*Bull. Entomol. Research*, 1914. Apr. Vol. 5. No. 1, pp. 49-60. With 4 plates and 1 map.
- (304) NEWSTEAD (R.) and DAVEY (J. B.). First Preliminary Report on the Bionomics of *Glossina morsitans*.—*Reports of the Sleeping Sickness Commission of the Royal Society*. 1914. No. 15, pp. 142-157. With 3 plates.

Helminths.

- (305) JOHNSTON (T. H.). Second Report on the Cestoda and Acanthocephala collected in Queensland.—*Ann. Trop. Med. & Parasit.*, 1914. Apr. 21. Vol. 8. No. 1, pp. 105-112. With 1 plate.
- (306) NICOLL (W.). The Trematode Parasites of North Queensland. II. Parasites of Birds.—*Parasitology*, 1914. June. Vol. 7. No. 2, pp. 105-126. With 2 plates comprising 13 figures.

Protozoa

- (307) DELANOË (P.) & DELANOË (Mme.). De la Rareté de *Pneumocystis Carinii* chez les Cobayes de la Région de Paris. Absence de kystes chez d'autres Animaux (Lapin, Grenouille, 3 Anguilles). [The infrequent Occurrence of *Pneumocystis carinii* in Guinea-pigs in the Neighbourhood of Paris. Absence of Cysts in other Species (Rabbit, Frog, and 3 Eels).]—*Bull. Soc. Path. Exot.*, 1914. Apr. Vol. 7. No. 4, pp. 271-272.
- (308) LEGER (M.) & LEGER (A.). Hématozoaires des Reptiles du Haut-Sénégal-Niger. [Haematozoa of Reptiles in Upper Senegal and Niger.]—*Bull. Soc. Path. Exot.*, 1914. June. Vol. 7. No. 6, pp. 488-493. With 6 text figures.

- (309) MINE (N.). Beiträge zur Kenntnis der Blutparasiten der Vögel in Japan. [The Blood Parasites of Birds in Japan.]—*Arch. f. Protistenkunde*, 1914. May 14. Vol. 34. No. 2, pp. 198–211. With 2 plates in colour comprising 109 figures.
- (310) MOLDOVAN (J.). Untersuchungen über den Zeugungskreis des *Leucocytozoon ziemanni* (Laveran). [Investigations regarding the Developmental Cycle of *Leucocytozoon ziemanni*.]—*Arch. f. Protistenkunde*, 1914. June 17. Vol. 34. No. 3, pp. 249–262. With 2 plates.
- (311) SCHEFERLING. Blutkörperchen und Blutparasiten. [Blood Corpuscles and Blood Parasites.]—*Zeitschr. f. Veterinärkunde*, 1914. May. Vol. 26. No. 5, pp. 214–219.

Unclassed.

- (312) CARPANO (Matteo). Su di alcuni Spirocheti rinvenuti in Neoformazioni papillomatose degli Equini. [Certain Spirochaetes met with in Papillomatous Neoplasms in Horses.]—*Annali d'Igiene Sperimentale*, 1914. Vol. 24 (Nuova Serie). No. 2, pp. 275–283. With 1 plate and 1 fig.
- (313) CARPANO (Matteo). Die nekrotisch-gangränösen Affektionen in der Veterinärpathologie. Die fuso-spirilläre Symbiose. [Necrotic and Gangrenous Affections in Veterinary Pathology. The Pseudo-spirillar Symbiosis.]—*Centralbl. f. Bakt.*, 1. Abt. Orig. 1914. June 13. Vol. 74. No. 3–4, pp. 225–241. With 1 plate.
- (314) CLANDON. Sur certaines Maladies du Mouton au Maroc. [Certain Diseases of the Sheep in Morocco.]—*Receuil. Méd. Vét.*, 1914. Apr. 30. Vol. 91. No. 8, pp. 151–155.
- (315) GROSSO (G.). Pathologisch-anatomische Veränderungen des Darmes und der Lunge des Affen (*Macacus*) durch tierische Parasiten verursacht. [Lesions in the Intestine and Lungs of the Monkey caused by Animal Parasites.]—*Zeitschr. f. Infektionskrankh. parasit. Krankh. u. Hyg. d. Haust.*, 1914. May 8. Vol. 15. Nos. 3–4, pp. 261–267. With 2 plates comprising 7 figures and 4 text-figures.
- (316) MEISSNER (W.). Das Kamelgestüt Kalkfontein-Nord der Kaiserlichen Schutztruppe in Deutsch-Südwestafrika. [The Kalkfontein-Nord Camel Stud of the Imperial Defence Force in German South West Africa.]—*Zeitschr. f. Veterinärkunde*, 1914. Apr. Vol. 26. No. 4, pp. 161–168.
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TROPICAL DISEASES BUREAU.

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[No. 4.]

BABESIASIS (PIROPLASMOSIS) AND ANAPLASMOSIS.

- (317) LAVERAN (A.) & FRANCHINI (G.). Contribution à l'Étude des "Marginal Points" des Hématies des Mammifères. [Contribution to the Study of "Marginal Points" in the Blood Corpuscles of Mammals.]—*Bull. Soc. Path. Exot.* 1914. July. Vol. 7. No. 7. pp. 580-584.

This paper contains a brief resumé of the references in literature to marginal points and to bodies having a close morphological resemblance to them.

The authors' observations have been carried out upon the blood of young animals of different species, and they have been able to find bodies having the morphological characters of marginal points in a number of them.

The blood of ten white mice weighing from 2 to 10 grammes was examined. In four of these anaplasma-like bodies, which the authors definitely describe as anaplasms, were not rare, and in the remainder they were present in small numbers. They were found in very small numbers in the blood of one of three adult white mice.

The authors have also discovered similar bodies in young grey mice and white rats, and in the blood of young rabbits and guinea-pigs, moles, cats, dogs, calves, goats, pigs, donkeys, and monkeys.

The morphology and staining reactions of the bodies are briefly described.

The authors conclude that bodies morphologically identical with those described by THEILER occur in the blood of young mammals apart from any infection, and that it does not appear to be possible to consider these as being of a parasitic nature. But it is said that, from the observations of SMITH and KILBORNE, and a number of other authors, it is possible that anaplasma-like bodies may represent a stage in the development of certain piroplasms.

- (318) CARPANO (M.). *Le Recidive nella Piroplasmosi. Su di un Tipico Caso di Recidiva nell'Asino.* [Relapses in Piroplasmosis. A Typical Case of Relapse in the Donkey.]—*La Clinica Veterinaria*. 1914. July 15. Vol. 37. No. 13. pp. 535-542. With 1 chart and 1 text fig.

In some of his previous papers the author has called special attention to the presence of latent forms of the parasite which occur in the blood of animals, and especially horses, affected with piroplasmosis, these forms having the morphological characters of anaplasms. They become active when for any reason the vitality and resistance of the host is lowered. A donkey which failed to become infected when inoculated subcutaneously with blood containing a small number of *Babesia caballi*, and also when subsequently inoculated intravenously with blood containing large numbers of *Nuttallia equi*, and was therefore immune to these parasites, had a relapse following an intravenous injection of streptococcic endotoxin derived from a very virulent strain.

Parasites of the *Nuttallia* type were found in the blood on the 12th and 13th days after the inoculation, and there was some amount of systemic disturbance.

The animal had been at the laboratory for 14 months before the injection which gave rise to the relapse was administered, and during that time it had not been exposed to infection by means of ticks.

- (319) MARTINEZ (I. G.). *Canine Babesiasis in Porto Rico.*—*Jl. Trop. Med. & Hyg.* 1914. July 1. Vol. 17. No. 13. p. 194.

Martinez briefly records the discovery of *Babesia canis* in the blood of dogs during the routine examinations for epizootic diseases and for rabies.

- (320) GOODALL (A.). *The Trypanblue Treatment in Piroplasmosis of Domesticated Animals in South Africa.*—*Parasitology*. 1914. May. Vol. 7. No. 1. pp. 62-68.

The notes contained in this paper are the outcome of the writer's experience of the use of the drug under practical conditions in country districts.

The solution used in every case was a 2 per cent. solution, made up with rain water, filtered and sterilised.

For dogs the dose used varied from 5 to 30 cc. according to the patient's weight, and was given subcutaneously.

Horses and large mules received 200 cc.; donkeys, ponies and small mules, 150 cc.; and cattle, 50 to 250 cc. In each of the latter cases the drug was given intravenously.

Good results have been obtained with all the species mentioned, and details are given of a few cases in each species. Conclusions:—

" 1. Parasites are not always present in the peripheral blood of dogs, and are often extremely difficult to find in other animals, especially under conditions encountered in the field. If well-marked clinical symptoms are shown one is justified in using Trypanblue, even if parasites cannot be satisfactorily demonstrated.

"2. A large dose of a weak solution of Trypanblue should be administered. The writer cannot help thinking that the failures attributed to the drug by other observers are due, in many cases, to too small a quantity of the solution being injected.

"3. After an experience of the treatment in some hundreds of cases, the writer feels justified in stating that animals do not suffer from relapses if a sufficient quantity of the solution is used.

"4. The dog can become infected a second time after Trypanblue treatment, but this second infection is also amenable to the drug."

TRYPANOSOMIASIS.

- (321) BRUCE (D.), HAMERTON (A. E.), WATSON (D. P.) & Lady BRUCE. **Trypanosome Diseases of Domestic Stock in Nyasaland. III.—*Trypanosoma pecorum*. Development in *Glossina morsitans*.**—*Proc. Roy. Soc.* 1914. Aug. 6. Series B. Vol. 88. No. B 600. pp. 33-37. With 1 coloured plate.

In a previous paper the authors have described the morphology and pathogenic range of this trypanosome, and in the present paper an account is given of its development in *G. morsitans*.

The trypanosome belongs to the group which does not invade the salivary glands. The development first takes place in the gut, and then extends forwards to the proboscis and hypopharynx, where the development into "blood forms" occurs.

Seven transmission experiments were carried out, and of these five were positive. Two of these were at laboratory temperature, and in the remainder the flies were kept in an incubator. Ten per cent. of the flies used were found to be infected.

From the details given it appears that a period of from 19 to 53 days may elapse before flies become infective.

In the positive experiments the majority of the flies showed infection of the proboscis, and all showed infection of one or more portions of the intestine. The salivary glands were always negative.

In the negative experiments there was infection of the labial cavity of one only. One or more parts of the intestine showed infection in every case, but the salivary glands were uniformly negative.

The coloured plate shows the cycle of developmental forms of *T. pecorum* in *G. morsitans*. In the intestine long parasites which are indistinguishable from the corresponding forms of other pathogenic parasites occur. When attached to the labrum the crithidial form is assumed, and in the proboscis the "blood forms," which are the only infective form, are developed.

- (322) BRUCE (D.), HAMERTON (A. E.), WATSON (D. P.) & Lady BRUCE. ***Glossina brevipalpis* as a Carrier of Trypanosome Disease in Nyasaland.**—*Proc. Roy. Soc.* 1914. Aug. 6. Series B. Vol. 88. No. B 600. pp. 20-32. With 1 coloured plate.

G. brevipalpis is found in a tract of country estimated at about 5 square miles at the mouth of the Lingadzi river on the west shore of Lake Nyasa. This area is traversed by two main roads, one running

east and west and the other parallel to the shore and about one mile west of it.

During the day no specimens of the fly are ever seen, but as evening approaches specimens may be seen sitting motionless in the road. They do not settle upon or follow passers-by, and would take no notice of a dog repeatedly walked through their haunts towards evening. They do not move about in search of food, nor do they chase each other.

Five hundred flies were caught and every one was a male. Some of them were found on dissection to contain mammalian blood. When kept in captivity they remain dormant on the sides of their cages during the day, but become active at night. If the side of the cage be placed up against a dog or a goat they will feed with avidity at any time during the day or night.

Examinations made to ascertain the natural food shewed that of 59 flies examined seven contained blood. In six cases it was probably that of antelope and in the other human.

Of the several hundred flies dissected, only four were found to be females, and it would appear that while the males are found on the paths the females remain in the jungle. The same kind of thing was observed to a lesser extent with *G. morsitans*.

Dissections of wild flies shewed that 44 out of 496 were infected with trypanosomes.

While trypanosomes were found in the proboscis, and in one instance in the salivary glands, the great bulk of them occurred in the mid and fore-gut, and in the smaller number of cases in the hind gut.

It is stated that in 19 cases it was possible to make a more or less correct guess at the identity of the trypanosomes found in the flies. These are *T. brucei* vel *rhodesiense*, 1; *T. pecorum*, 9; *T. simiae*, 1; and *T. grayi*, 8. In 10 flies the flagellates were considered to belong to a pathogenic type of unknown species, and in the remainder no opinion could be expressed.

Five experiments were made with flies brought from the lake shore; in each case a monkey, a dog, and a goat being used for feeding experiments. In only one case was the result positive, *T. pecorum* being found.

A number of experiments were carried out to ascertain whether *G. brevipalpis* is capable of acting as a carrier of various pathogenic trypanosomes.

T. brucei vel *rhodesiense*.—Wild flies were used in the experiments, as it was found to be impossible to breed flies for the purpose, both on account of the distance that they had to be carried from the lake to the laboratory (50 miles), and on account of the scarcity of the flies.

Wild flies were fed upon animals infected with the trypanosome causing disease in man in Nyasaland and then upon healthy animals. In six experiments 232 flies were used, but the result was negative in every instance. Fifty-three flies were dissected and eight infected flies were found. In one of these there was infection of the proboscis and alimentary tract, in one infection of the salivary gland and of the alimentary tract, and in the remainder infection of the alimentary tract only. The two flies in which there was infection other than

that of the intestine were found in one batch. That in which the salivary glands were found to be infected was dissected at the 33rd day, and the other at the 46th day. The former of these had the salivary glands swarming with trypanosomes, and, although inoculation of a rat failed, it is thought probable that it represented a true development of the Nyasaland trypanosome, the development not having reached the infective stage.

In a coloured plate are shewn the morphological details of the trypanosomes occurring in the fly.

In the intestine of the fly long slender forms are found, and these invade the salivary glands. Here the parasite acquires a crithidial form, in which the posterior extremity is attenuated while the anterior end becomes broader. Later the posterior portion becomes short again and the parasite is short and thick. The parasite then becomes rounded in form, and masses of them in this condition are found in the lumen of the glands. These masses break up into individuals in which the micronucleus is again posterior and the parasite gradually assumes the fully developed trypanosome, or "blood form." It is pointed out that although wild flies were used, this is the only type of trypanosome which invades the salivary glands of the flies in the district and that therefore *T. pecorum*, *T. simiae*, and *T. caprae* are excluded.

T. brucei Zululand, 1913.—Three experiments were carried out, using 200 flies. One was positive and two negative.

Only 12 flies were dissected, and in one long trypanosomes apparently of the pathogenic type were found in the intestine, but it was impossible to identify them. A dog became infected 58 days after the flies had been fed upon an infected dog. Only two flies of this batch were dissected, and both were negative. It is probable that the dog was infected by a fly in which development of *T. brucei*, Zululand, had taken place, as if there had been a fly in the cage naturally infected with the Nyasaland strain, the dog ought to have shewn trypanosomes earlier than the 58th day.

Further experiments of the same kind shewed that *G. brevipalpis* is capable of acting as a carrier for both *T. pecorum* and *T. caprae*. The flies dissected in these experiments shewed trypanosomes in the proboscis and intestine, but not in the salivary glands.

(323) CHALMERS (A. J.) & O'FARRELL (W. R.). Sleeping Sickness in the Lado of the Anglo-Egyptian Sudan.—*Jl. Trop. Med. & Hyg.* 1914. Sept. 15. Vol. 17. No. 18. pp. 273-284. With 1 map and 5 charts.

In this paper are compared *T. rhodesiense* and the Yei strain of trypanosomes. The former was brought from Liverpool and the latter was obtained from patients at Yei in the Lado district of the Mongalla Province.

The experimental work was done at Khartoum owing to the possibility of introducing *T. rhodesiense* into the sleeping sickness area. The patients infected with the Yei strain were not brought to Khartoum for examination because of the danger of spreading the disease, and because it was not desired to interfere with their treatment.

All the blood films were fixed wet with osmic acid vapour for about four seconds and then transferred to absolute alcohol, in which they

were kept for two to five minutes. Giemsa's stain was used in two dilutions: either 1 drop to 1 cc. with the addition of a drop or two of 1 per 1,000 potassium carbonate, in which they were stained for an hour or longer, or 2.5 cc. to 100 with 5 drops of carbonate solution, in which they were stained for 5 to 24 hours.

A summary of the state of affairs regarding African human trypanosomes and an interesting account of the history of the occurrence of sleeping sickness in the countries adjoining the Lado are given.

From these it appears that there are two main areas of infection in the Mongalla Province, namely a western area centered round Yei, and an eastern area adjoining Kajo-Kaji.

The Yei strain was obtained from a monkey inoculated by RANKEN from a patient at the Sleeping Sickness Segregation Camp at Yei.

It would appear that these two districts obtained their infection originally from two different sources. The first appears to have been infected from the Belgian Congo and possibly through the medium of Baganda traders. The source of the infection of the second district appears to have been Uganda.

The infected animals, two monkeys and a dog, were inoculated from an untreated case.

The authors state that the strain may represent the strain present in Uganda or that on the Congo, but they consider this to be of little importance as they believe that the Uganda infection was originally derived from the Congo.

Morphology.—The parasite varied in length from 18 to 36 microns, and in width from 1 to 2.5 microns. The average length of 1,000 trypanosomes, measured 100 daily, in the blood of an infected monkey was 25 microns. Measuring was commenced on the first day that the animal shewed a heavy infection, viz., the 10th, and it was continued to the 19th day. The monkey died on the 33rd day. A posterior-nuclear form was never observed.

Animal experimentation shewed that the virulence of the trypanosome for dogs, cats, rabbits, gerbils, jerboas, white rats, and monkeys, is less than that of *T. rhodesiense*, but greater than that of *T. nigeriense* Scott-Macfie.

In the dog the period of incubation was 12 days, and the animal often lived several months. In the monkey the period of incubation was nine days, and the duration of life averaged about 35 days. In the gerbil the period of incubation was 7 days, and the duration of life 14.

A dog was rendered immune, i.e., its peripheral blood failed to shew trypanosomes for more than 60 days after its fifth inoculation with the Yei strain. The last tested gerbil inoculated with the blood of this dog failed to become infected.

The serum of this dog destroyed the Yei trypanosome in vitro in 20 minutes, but was without effect upon *T. rhodesiense* after an hour.

At a time when the immunity was incomplete the serum was mixed with the different strains of trypanosomes and immediately inoculated into gerbils. In the case of the Yei strain an infection resulted, and the animal was recovering from it when it died of heat stroke along with 49 other animals. This occurred on the 39th day. The *rhodesiense* mixture resulted in an infection which proved fatal on the 7th day.

When the immunity was complete the serum was mixed with infected blood in the proportion of 10 to 1, and after 30 minutes was injected into gerbils. No trypanosomes were seen in the blood of the gerbil inoculated with the Yei strain, but it was killed accidentally on the 13th day after inoculation. No trypanosomes were found in the internal organs, but smears from the lungs shewed peculiar bodies contained in cells which resembled the granules found in the spleens of patients and of animals inoculated with kala azar. With *T. rhodesiense* the gerbil died of infection on the 10th day.

Cross immunity is to be dealt with in a subsequent paper.

Cytolytic sera.—Human serum was not found to have any trypanolytic effect upon either of the human strains of trypanosomes or upon two strains of mule trypanosomes, but it did appear to have a distinctly beneficial effect upon the Yei strain, as after an hour the trypanosomes were more active than at the commencement of the observation.

It is recorded that a strong though incomplete agglutination was observed to be produced by a normal human serum upon a mule strain of trypanosomes, and it is pointed out that if this is admitted agglutination is useless as a specific test.

No experiments have been carried out regarding the transmission of the trypanosomes.

A detailed comparison is made with each of the other human trypanosomes, excluding the original strain of DUTTON, 1902, which the authors have not been able to trace.

The conclusions arrived at may be summarised as follows :—

The strain sent in by RANKEN and the Congo strain are the same, and probably they and the Uganda strains are identical. There being no data of which the authors are aware to compare these strains with *T. gambiense*, Dutton, 1902, they think that provisionally it would be safer to keep the name *T. castellanii*, Kruse, 1903, for these strains until more light is thrown upon the question as to what is *T. gambiense*, Dutton, 1902.

It appears that sleeping sickness in Africa can be divided into three categories.

(1) Southern sleeping sickness caused by *T. rhodesiense* and spread by *G. morsitans*.

(2) Equatorial sleeping sickness caused by *T. castellanii*, Kruse, 1903, and spread by *G. palpalis*.

(3) Northern sleeping sickness which may be caused by as yet imperfectly known trypanosomes, named :—

(a) *T. gambiense*, Dutton, 1902.

(b) *T. nigeriense*, Scott-Macfie, 1913, and possibly other trypanosomes.

The authors believe that BRUCE's method of charting trypanosomes and LAVERAN and MESNIL's method of cross-immunity testing are both of distinct value in the comparison of parasites.

The question as to why some cases of the disease in the Lado Enclave are exceedingly chronic while others are very acute may be answered in two ways. The more probable reason is that the chronic cases occur in the area that has been longest infected, while the acute cases occur in the area which has recently become infected.

The less likely answer is that there are two forms of the disease.

- (324) MESNIL (F.) & RINGENBACH (J.). Sur le *Trypanosoma rhodesiense* et ses Affinités avec le *Tr. gambiense*. Quatrième Note. [*Trypanosoma rhodesiense* and its Affinities with *T. gambiense*. Fourth Note.]—Bull. Soc. Path. Exot. 1914. July. Vol. 7. No. 7. pp. 612-618.

The authors refer to a paper published by them in 1911 in which it was recorded that a monkey which had a strong immunity to two strains of *T. gambiense* subsequently became infected with *T. rhodesiense* although the period of incubation and the duration of life were longer than in monkeys having no immunity to *T. gambiense*.

Reference is also made to LAVERAN's experiment in which a goat immune to *T. gambiense* contracted a fatal infection which lasted only 35 days when inoculated with *T. rhodesiense*.

The conclusion is drawn from these results and from those of MESNIL and LEGER, in which mice inoculated with *T. rhodesiense* and cured with arsenophenylglycin were tested as to their susceptibility to *T. gambiense*, that the two viruses constitute distinct but closely related species.

The present paper records experiments carried out with a strain of *T. rhodesiense* (Case 16, W. G.) obtained from WENYON, and the strain originally studied by the authors in 1911-12.

In the opinion of the authors, negative results obtained with cross immunity experiments are less conclusive than positive ones.

In the rat WENYON found that the duration of life varied from 12 to 39 days, but in the series of passages through the rat carried out by the authors the period ranged from five to 18 days only.

In the mouse the course of the infection is still more variable than in the rat. The duration of life varied from four to 25 days, and in cases in which the animals survived for more than 15 days crises occurred during which trypanosomes were not discoverable in the blood by microscopical examination. In the course of the later passages the duration of life was longer than in the earlier ones.

A similar variability of effect was also observed in the guinea-pig.

The authors' own strain, on the other hand, was markedly constant in its effects. Mice were regularly killed in four days.

Guinea-pigs died in 28 and 22 days, as compared with 19 and 79 days with WENYON's strain.

A series of experiments was carried out with four goats, two of which were immune to *T. gambiense* and two susceptible. Each of the strains of *T. rhodesiense* was used to inoculate an immune and a susceptible goat.

Goat B. immune to *T. gambiense*.—Inoculated with the authors' strain of *T. rhodesiense* became infected and died. Trypanosomes appeared in the blood in small numbers a month after inoculation, and they were observed on a number of occasions afterwards, but apparently always in small numbers. The goat died six weeks after the appearance of trypanosomes in the blood. There was oedema of the face and simple keratitis. Duration of life, 68 days.

Goat B¹. Non-immune to *T. gambiense*.—Infection resulted, but trypanosomes were not numerous in the blood. There was slight oedema of the face, but there is no mention of keratitis. The animal died in 42 days.

Goat C. Immune to *T. gambiense*.—Inoculated with WENYON's strain. Although a single trypanosome was seen in the blood on one occasion only about seven weeks after inoculation, the infectivity of the blood was repeatedly proved by the inoculation of mice. The animal died on the 54th day.

Goat C'.—Non-immune goat inoculated with WENYON's strain of *T. rhodesiense*. Microscopic examination of the blood was negative throughout, but its infectivity was proved by the inoculation of mice from time to time. The animal died on the 61st day.

It is concluded from these experiments that the evidence tends to show that *T. rhodesiense* and *T. gambiense* are distinct.

A few serum experiments were carried out, and it was found that the sera of the two non-immune goats possessed no activity against *T. gambiense*. The sera of Goats B and B' were found to be active against *T. rhodesiense* of the authors' strain, but to be practically without effect upon WENYON's strain. The sera of Goats C and C' were found to be fairly active against WENYON's strain, but very slightly against the authors' strain.

In 1911 the authors stated that their strain of *T. rhodesiense* behaved like an animal trypanosome of the nagana type from the point of view of the action of human serum upon it. WENYON's strain was found to have practically the same property.

(325) BRUCE (D.), HAMERTON (A. E.), WATSON (D. P.) & Lady BRUCE. Infectivity of *Glossina morsitans* in Nyasaland during 1912 and 1913.—*Proc. Roy. Soc.* 1914. Aug. 6. Series B. Vol. 88. No. B 600. pp. 43-48.

During 1912, 1,975 flies were caught and dissected. Of these 86 per cent. were males and 14 per cent. females. 6·53 per cent. were found to be infected with trypanosomes. In 1913, 1,060 flies were dissected, and 8·58 were found to be infected.

During 1912, 60 attempts were made to determine the infectivity of the flies by injecting the intestinal contents into animals, but only three inoculations were positive. This was no doubt to a large extent due to the fact that the intestinal forms are not infective, a point subsequently discovered. In only two cases were the salivary glands found to be infected. Inoculation of these glands into rats confirmed the diagnosis that the infection was *T. brucei* vel *rhodesiense*.

In 1913, an attempt was made to diagnose directly the species of trypanosome with which the fly were infected, and no injections were made.

Invasion of the salivary glands could only be *T. brucei* vel *rhodesiense*; invasion of the intestine, labial cavity and hypopharynx meant *T. pecorum* or *T. simiae*, and size would distinguish between these. Lastly, if only the labial cavity and hypopharynx were seen to contain flagellates, *T. caprae* was indicated.

T. brucei vel *rhodesiense* was found once, *T. pecorum* six times, *T. simiae* 12 times, and *T. caprae* 14 times.

- (326) BRUCE (D.), HAMERTON (A. E.), WATSON (D. P.) & Lady BRUCE.
Trypanosomes found in Wild *Glossina moritans* and Wild Game in
the "Fly-Belt" of the Upper Shiré Valley.—*Proc. Roy. Soc.* 1914.
Aug. 6. Series B. Vol. 88. No. B 600. pp. 38-41.

The "fly-belt" in question extends along the Upper Shiré River Valley from Lake Pamalombe to the Murchison cataracts. It is about 100 miles south of Kasu. It is separated from the "fly" area on the west shore of Lake Nyasa by a range of hills where fly is absent, but which forms no obstacle to the passage of infected animals from one area to the other.

Game is abundant in the district, particularly in the dry season. Cases of trypanosomiasis have been observed in man, and the natives are unable to keep cattle. Their goats and dogs are constantly being destroyed by trypanosomiasis.

Seven experiments were carried out, but only dogs and goats were used, as there were no monkeys available. The different batches of flies, which numbered from 73 to 650 in a batch, were allowed to feed upon a dog and a goat twice.

Six of the dogs became infected with *Trypanosoma brucei* vel *rhodesiense*. Two of these were at the same time infected with *T. pecorum*, and one contracted a *pecorum* infection alone. These were the only trypanosomes found in the dogs.

Six of the goats became infected with *T. pecorum*, one of these also had *T. simiae*, four were infected with *T. caprae*, and one showed a *caprae* infection only.

These experiments show that the fly in the Upper Shiré district carries the same species of trypanosomes as those found in the flies from the Proclaimed Area.

A zebra, 7 impala, 2 koodoo, and 6 waterbuck were examined for trypanosomes, the examination involving the microscopic examination of blood preparations. *T. brucei* vel *rhodesiense* was found in one waterbuck, *T. pecorum* was found in 2 impala and 2 waterbuck, *T. caprae* was found in an impala, a koodoo, and a waterbuck, and *T. ingens* was found in one waterbuck.

- (327) BRUCE (D.), HAMERTON (A. E.), WATSON (D. P.) & Lady BRUCE.
The Food of *Glossina morsitans*.—*Proc. Roy. Soc.* 1914. Aug. 6.
Series B. Vol. 88. No. B 600. pp. 41-42.

Five hundred flies, freshly caught in the Proclaimed Area, were killed by chloroform and a smear made from the intestine of each. These were fixed with osmic acid and alcohol, stained with Giemsa, and examined for the presence of blood corpuscles.

The proportion of male to females was roughly two to one.

Of the 500 flies examined, 57.6 per cent. were found to contain mammalian blood in a recognisable state. No measurements of the corpuscles were made owing to changes in the corpuscles induced by digestion, but cells of the small type such as are seen in the blood of antelope predominated.

In three cases nucleated red corpuscles were found, and in one of these only were there no mammalian corpuscles present at the same time. Measurement of these corpuscles showed that they were in all

probability avian and not reptilian. No vegetable matter was found in any of the flies.

From experiments carried out with laboratory flies it appeared that blood was not recognisable after the third day. Hence it may be inferred roughly that half the flies examined had fed within three days, and that therefore flies naturally feed at least once in six days.

Trypanosomes were recognised in 2·8 per cent. of the flies, and it is probable that a higher percentage was actually infected.

(328) OWEN (G. E.). **Mechanical Transmission of Trypanosomiasis.**—*Jl. Comp. Path. & Therap.* 1914. Sept. Vol. 27. No. 3. pp. 259–260.

The author's object in writing this paper is to point out the possibility of mechanical transmission becoming serious under suitable conditions, rather than to lay stress upon its frequency and importance.

During the period 1908–1913, cattle to the number of 2,500–3,000 died from trypanosomiasis in the Barotse reserve of Northern Rhodesia, although the district is free from *Glossina morsitans*. Special interest attaches to the fact that the mortality began each year about the beginning of February and ceased after June. The wet season, when flies are most numerous, lasts from December to March.

There were in the district some dozen horses, some being bred every year, and large numbers of sheep, dogs and goats, but no natural case of trypanosomiasis had ever been encountered among these animals. The explanation of this fact did not lie in the trypanosome being pathogenic for cattle only, as the author found all the other species susceptible to it, and particularly the dogs.

In seeking an explanation of these facts the first point of importance is that the trypanosomiasis was definitely confined to certain herds, others grazing over the same ground being free, and in every native herd there was a history of exchanging cattle with, or hiring to traders and transport riders.

In 1908, an owner came into the district with sick cattle. He lost the majority of them from trypanosomiasis in three years, and with his advent the disease became very noticeable.

In 1912, when the writer went to investigate, he at once resorted to isolation, with the result that in 1913 he had over 800 head of cattle without loss in an area which had been the centre of the disease the previous year. These animals remained out of a lot of 1,100. They were carefully watched for about six months after the fly season, all infected and suspected animals being removed before the flies became numerous again.

During April and May, 1912, the author saw 200 cases, whereas from February to May inclusive 1913 only five cases were seen, all occurring in a suspected herd.

Had a fly or a tick been associated in the transmission, the disease could not have remained confined to certain herds while others on the same veld remained unaffected.

Tabanidae, Haematopota and Stomoxys are the commonest flies in the district.

During the months when the flies are most numerous and during the hot period of the day, the cattle crowd together for mutual pro-

tection. To watch a herd crowded together and see the Tabanidae swarming on them is sufficient to impress one with the fact that the conditions are very favourable for mechanical transmission.

The following experiment was made :—

Two kraals were erected within 10 yards of each other. In one two healthy cows and a heifer were placed, and in the other two healthy cows and two infected cows. The cattle from the two kraals were grazed over the same ground, but never at the same time, and the kraals were occasionally changed.

The two healthy cattle in the second kraal became infected, but the healthy animals in the other kraal remained uninfected. The object of exchanging the kraals was to ascertain whether mosquitoes in the kraals would act as transmitters.

Dogs, sheep, and goats were exposed to infection by mixing healthy with clean, but no infection resulted.

It was noticed that while dogs allowed Stomoxys to feed upon them freely, they bit at any Tabanidae coming near them. No positive results were obtained with experiments carried out to test cyclical or mechanical transmission with either of these species.

The author came to the conclusion that the trypanosome, which was not identified, but was responsible for a chronic infection in the cattle, was transmitted in a mechanical manner by Tabanidae.

(329) CROSS (H. E.). *Experiments on the Treatment of Surra in Camels.*—*Memoirs Dept. Agric., India.* Veterinary Series. 1914. Sept. Vol. 2. No. 6. pp. 155–198. With 2 figs.

This paper contains an account of the experiments carried out with the following six methods of treatment :—*

1. One dose of soamin followed by gradually increasing doses of arsenious acid ; the treatment being commenced when the trypanosomes were present in the peripheral blood.

2. As above, but treatment started when the trypanosomes were absent from the circulation.

3. Gradually increasing doses of arsenious acid ; treatment being started when the trypanosomes were present in the blood.

4. As No. 3, but started when trypanosomes were absent from the peripheral blood.

5. Combination of soamin, tartar emetic, and arsenious acid ; treatment being started when trypanosomes were present in the blood.

6. Soamin and arsenious acid given alternately, with one day interval between each ; treatment being started when trypanosomes were present in the blood.

As a preliminary investigation a number of experiments were made to ascertain the minimum lethal dose of arsenious acid when administered intravenously.

*Reference to the list of treatments given at the commencement of the paper appears to indicate that there has been a little confusion in the numbering of treatments 3 and 4, as in the detailed statements of the experiments under these numbers the factor regarding the presence or absence of trypanosomes at the time when treatment was started appears to have been reversed. The numbering given in the original has been followed in this abstract.

The tolerance of the camel for arsenious acid is very variable and consequently each case has to be treated upon its merits to some extent. It was found, however, that as a general rule 1·2 grammes per 1,000 lb. body weight can be safely given, but in some cases larger doses (1·7 gm.) were tolerated.

The arsenious acid was used in every case in a 1 per cent. solution dissolved in alkali. Tartar emetic was also administered in a 1 per cent. solution. The solutions were filtered and injected at 37° C. by means of Ehrlich's injection flasks.

In treating cases of colic large doses of chlorodyne (3 to 4 ounces), containing 1 grain of hydrochloride of morphia to the ounce, should be administered. In cases of unsteadiness 2 grains of strychnine should be given subcutaneously.

A liberal diet should be allowed during and after treatment.

Treatment 1.—The dose of soamin used was 100 cc. of a 5 per cent. solution and the dose of arsenic ranged from 0·4 gm. to, in some cases, over 2 gm., about 10 doses of arsenic being given, and one day elapsing between consecutive doses.

In all, 21 animals were treated. In 12 cases there were relapses, in two cases the animals died, and in the remaining seven cases there was no relapse recorded for periods varying from 111 to 281 days. The percentage of animals in which no relapse was recorded was 33.

Treatment 2.—Nine animals were subjected to this method of treatment. In two cases there were relapses, in two instances the animals died, and in the remaining five no relapses were recorded for periods varying from 95 to 281 days.

In this case 55 per cent. of the animals did not show any relapses.

Treatment 3.—Arsenious acid alone was administered in doses increasing from 0·4 gm. to 2 gm. or more, there being, as before, an interval of a day between consecutive doses.

The following table summarises the results obtained by this method :

| | |
|---------------------------------------|--|
| Number of animals treated .. | 12 |
| Cases in which relapses occurred .. | 4 |
| Number of Deaths | 1 |
| Cases in which no relapse occurred .. | 7 (122-277 days under observation). |

Treatment 4. The same as No. 3, but started when trypanosomes were present in the blood.—

Eight animals were treated by this method. In four cases there were relapses, and in the remaining four no relapses were recorded for periods ranging from 121-178 days.

Treatment 5. Combined soamin, tartar emetic, and arsenious acid.—

These drugs were given on three consecutive days and then repeated three times with intervals of two to six days. The doses of soamin and emetic was 100 cc. of a 5 per cent. solution (and in a few cases 6 or 7 per cent.) and 0·5 gm. respectively. The dose of arsenic was gradually increased from 0·5 to 1·2 gm.

The tabulated results are as follows :—

| | |
|---------------------------------------|---|
| Animals treated | 24 |
| Cases in which relapses occurred .. | 9 |
| Number of Deaths | 1 |
| Cases in which no relapse occurred .. | 14 (73 to 379 days under observation.) |

Percentage of cases in which no relapses were recorded—58 per cent.

Treatment 6. Soamin and arsenious acid given alternately.—The soamin was given in a 5 to 7 per cent. solution, and the arsenious acid in doses increasing up to a maximum of 1·5 grammes. Included among these experiments are some in which the treatment was continued by arsenious acid alone after some doses of soamin had been administered.

Fifteen animals were included in this group. In four cases there were relapses. In one death occurred, and in the remaining 10 (66 per cent.) no relapses were recorded in from 80 to 279 days.

The conclusions drawn may be summarised as follows :—

There is no necessity to wait for the appearance of trypanosomes in the circulation before commencing treatment. This is of importance because sometimes the intervals between paroxysms last for weeks and time is thus saved.

No case should be considered as cured until it has been under observation for a year without a relapse occurring.

Soamin should not be used in solutions stronger than 5 per cent. as there is a danger of producing nephritis. While cures can be effected without increasing the doses of arsenic to the subtoxic dose, the dose should be increased till the sub-tonic dose is reached, the principle of the treatment being the sterilisation of the tissues.

In the combined treatment, the dose of arsenic must not be so large as when it is given alone or when alternated with soamin.

As the tolerance of the camel to arsenic varies from animal to animal and the virulence of the trypanosome varied in different outbreaks, the same percentage of cures cannot always be obtained.

Good feeding is essential.

(330) CRUCA (M.). *L'Action de quelques Substances Médicamenteuses sur le Pouvoir Alexique du Sérum*. [The Effect of Certain Medicinal Substances upon the Alexic Power of Serum.]—*Bull. Soc. Path. Exot.* 1914. July. Vol. 7. No. 7. pp. 626-632.

The experiments detailed in this paper were carried out during the course of an investigation into the mechanism of the action of emetic upon trypanosomes.

Reference is made to the observations of FÉNYVESSY and FREUND, who found that while the intravenous injection of calcium chloride increased the alexic power of the serum, the opposite effect was produced when the drug was added to serum in vitro.

The observations of WEIL and DUFOURT and of WEISBACK are also referred to. These authors found respectively that the intravenous injection of 2 to 4 cc. of a 4 per cent. solution of bicarbonate of soda causes an increase in the alexic power of the serum, both in man and in rabbits, and that salvarsan produces an accelerating effect upon haemolysis in vitro and particularly upon the alexin.

So was unable to detect any influence upon the alexin when rabbits were injected subcutaneously with atoxyl.

The author's experiments have been carried out upon rabbits, guinea-pigs, and rats, both normal and infected with the Uganda strain of Nagana.

The experiments were carried out in the following way :

The rabbits and guinea-pigs were injected intravenously and the rats intraperitoneally with emetic in the proportion of 1 to 5 mgm. per 100 gm. body weight, atoxyl in doses up to 2 mgm. per 100 gm. and salvarsan 1.5 mgm. per 100 grammes.

The animals were either bled to death half an hour afterwards or they were bled on a number of occasions at variable intervals. The results of the experiments are set out in a number of tables.

It was found that the intravenous injection of emetic or salvarsan into healthy animals caused an elevation of the alexic power, while atoxyl produced the opposite results. The latter effect was far more marked than the opposite effect produced by emetic and salvarsan. That the effect was due to an action upon the alexin is shown by the fact that heating all the sera to 56° C. inactivated them. The effect could be observed for 48 hours after the injection.

With regard to animals severely affected with trypanosomiasis, it was confirmed that at the height of infection alexin is generally quite absent from the serum, but in these animals when treated with emetic or salvarsan the increase in the amount of alexin was particularly marked. Such sera were able to activate systems which could not be activated by sera from non-infected animals three hours after the treatment.

Atoxyl decreases the alexin in non-infected animals, but increases it in infected animals, but to not so great an extent as the other two drugs.

Among healthy rabbits and rats the alexin is sometimes particularly susceptible to the action of chemical salts.

In the earlier experiments the control serum was taken from an animal before the injection of the drug. The inconclusive and sometimes inconstant results were due to a diminution in the amount of alexin following on the previous bleeding.

In some of the experiments the alexin was found to have fallen to a quarter of the normal titre when an animal was bled twice on the same day or on two consecutive days.

The increase in the alexin is associated with and follows the intense leucocytosis produced by the injection of the drugs.

The fall in the amount of alexin produced in healthy animals by atoxyl is probably due to the affinity of this drug for the albumens of the body, this being essential to render it active (LEVADITI and YAMANOUCI). It is quite possible that in animals infected with trypanosomes the product which is formed in the body has an action comparable to that provoked by salvarsan and emetic.

(331) LAVERAN (A.) & ROUDSKY (D.). *Sur un Dérivé du Diamino-arsénobenzène*. [A Derivative of Diaminoarsenobenzene].—*Bull. Soc. Path. Exot.* 1914. July. Vol. 7. No. 7. pp. 593-596.

The substance described in this paper has been prepared by OECHSLIN and has been named O 1. It is said to possess the following advantages over arsenophenylglycin, which it closely resembles in its action in trypanosome diseases.—

The drug is far more stable. It occurs in the form of a pale yellow powder which is easily soluble in water. It is far cheaper to prepare and does not undergo rapid alterations when sealed up in tubes, even

TABLE GIVING THE SALIENT CHARACTERS OF THE MORE IMPORTANT MAMMALIAN TRYPANOSOMES

| MORPHOLOGY | | | | | | | | | | | | |
|----------------------------------|--|------------------------------------|--------------------------------------|-----------------------------|------------------------------|----------------------------------|--|--|--|--|--|--------------------------------|
| Qualitative Characters | | | | Quantitative Characters | | Pathogenicity | | Development in insect host when known | | Where first found | | Remarks |
| Flagellum | Flagelliphagoclast | Nucleus | Cytoplasm | Length in microns Min. Max. | Breadth in microns Min. Max. | Acute for all laboratory animals | | Unknown | | Zaireland India Central America North Africa South America Cameroon German East Africa Uganda Transvaal Transvaal Uganda N.E. Rhodesia (World wide) Brazil | | *Roubaud & Bouet's observation |
| Always free | | | | 18-34 | 1.5-2 | Acute for all laboratory animals | | Unknown | | Zaireland | | |
| " | | | | 18-34 | 1.5-2 | " " | | " " | | India | | |
| " | | | | 18-28 | 1.5-3 | " " | | " " | | Central America | | |
| " | | | | 16-35 | .. | " " | | " " | | North Africa | | |
| " | | | | Average 22 | 1.5 | " " | | Unknown | | South America | | |
| Absent | | | Club-shaped | 16-31 | 2-3 | Equidae and ruminants only | | Limited to the proboscis in glossina | | Cameroon | | |
| " | | | Heavier build than <i>vitax</i> | 18-32 | 1.75-4.25 | " " | | " " | | German East Africa | | |
| " | | | | 12-19 | 1.5-2.5 | " " | | " " | | Uganda | | |
| " | Near nucleus | | | 18-50 | 4-6 | Slight to bovidae only | | Unknown | | Transvaal | | |
| " | | | | 25-70 | 2-5 | " " | | Possibly Hippoboscids, details unknown | | Transvaal | | |
| " | Near nucleus | | <i>Myonemes</i> | 72-122 | 7-10 | Unknown | | Unknown | | Uganda | | |
| " | Near nucleus | | " | 52-72 | 5-8.5 | " " | | " " | | N.E. Rhodesia | | |
| " | Rod shaped, transverse | Junction of ant. and middle thirds | | Average 24 | .. | Rats only infected | | Instances of rat fleas and lice | | (World wide) | | |
| " | Large ovoid | | | Average 20 | .. | Slight for laboratory animals | | | | Brazil | | |
| Never free | | | | 8-19 | Average 1.5 | Acute for all laboratory animals | | Gut and proboscis of glossina | | Congo | | |
| " | | | | 10-16 | Average 1.5 | Equidae and ruminants only? | | " " | | Southern | | |
| " | | | Heavier build than <i>congolense</i> | 10-20 | Average 3.3 | Slight for laboratory animals | | Unknown | | N.E. Rhodesia | | |
| " | Frequently projects as lateral excrescence | | | 12-23 | .. | Acute for monkeys and pigs only | | Proboscis of <i>Glossina morsitans</i> | | New Zealand | | |
| Sometimes free and sometimes not | | | | 13-39 | .. | Acute for laboratory animals | | Gut and salivary glands of glossina | | Gambia | | |
| " | | | | 10-33 | .. | Slight for laboratory animals | | Unknown | | N.E. Rhodesia | | |
| " | | | | 8-37 | .. | " " | | " " | | Nigeria | | |
| " | | | | 14-35 | .. | Acute for laboratory animals | | Gut and proboscis of glossina * | | Senegal | | |
| " | | | | 12-39 | .. | " " | | Gut and salivary glands of glossina | | N.E. Rhodesia | | |
| " | | | | 13-38 | .. | " " | | " " | | Uganda | | |
| " | | | | 14-36 | .. | " " | | Transmitted by ectus | | North Africa | | |

* Roubaud & Bouvier-Lemerle

when the tubes contain a small amount of air. A watery solution is said to remain unchanged in sealed ampoules for periods varying from eight days to a month. It is less toxic than arsenophenylglycin. A healthy mouse weighing 20 grammes will withstand a dose of 10 mgm. whereas a dose of 8 mgm. of arsenophenylglycin would suffice to produce a fatal effect. As animals infected with trypanosomes are more susceptible to the action of drugs than healthy ones, the dose for an infected mouse must not exceed 5 mgm. For a guinea-pig weighing 500 grammes (whether infected or not is not stated) the dose is 5 cgm., and for a dog weighing 10 kgm., 25 cgm.

No danger attaches to repeated treatments with O 1. Subcutaneous injections produce painful oedematous swellings, and sometimes gangrene of the skin, but a 4 per cent. solution in distilled water has been injected into the saphena vein of dogs without ill effects.

Sterilisation in the autoclave increases the toxicity of the drug, and the authors have resorted to filtration for the sterilisation of solutions destined for intravenous injection.

The subcutaneous injection of 5 mgm. into mice heavily infected with *T. brucei*, *T. rhodesiense*, or *T. dimorphon* has caused the disappearance of the trypanosomes within a few hours. Cures have been effected by this treatment and also by three injections of 2 to 3 mgm.

In the dog and the guinea-pig it is easy to cause a disappearance of trypanosomes from the circulation, but the authors are not in a position to state whether relapses occur, and in what proportion of cases.

A cure has been effected in the case of a guinea-pig infected with a strain of *T. gambiense* resistant to atoxyl.

Neither the composition nor the method of preparation of the drug is given.

(332) LEVADITI (C.) & MUTERMILCH (St.). *Ambocepteurs et Arsénobenzol*. [Amboceptors and Arsenobenzol.]—*Bull. Soc. Path. Exot.* 1914. July. Vol. 7. No. 7. pp. 633-640.

The authors have prepared three kinds of sera in the following ways: (a) Serum from normal rabbits treated with "606." (b) Serum containing trypanolytic amboceptor from infected rabbits killed after the crisis. (c) Serum possessing the characters of the two preceding from infected rabbits which have been treated with "606" after the crisis.

These three sera were used for the treatment of infected mice.

Eight experiments have been carried out in this way, and in five of these constant results were obtained.

Comparison of the results obtained shews that what may be called the "606 alone" serum does not exercise any marked effect upon the multiplication of the trypanosomes, nor does it delay death very much. The serum containing only amboceptor exercised a very feeble effect, all the mice dying with trypanosomes in their blood. On the other hand the serum from the rabbits treated with "606" which also contained amboceptor prevented the multiplication of the parasites, the mice which received the largest doses of this serum shewing no

trypanosomes in their blood during the whole course of the experiments which lasted from 22 to 37 days.

In three of the experiments the results obtained were, however, different, in that the "606 alone" sera used in these experiments were themselves active, so that the beneficial effects of combining amboceptor with "606" were less apparent.

Having established the superiority of the sera combining the two active properties, the next step taken was to ascertain the trypanocidal action of the three sera *in vitro*.

The experiments were repeated several times with constant results. Two striking facts were ascertained.

(1) If the trypanocidal effect of the amboceptor + "606" complex is compared *in vitro* with the trypanocidal action of each of the components, the effect of the former is not found to greatly exceed the effects produced by the latter. Thus, in one instance the complex was found to be completely effective in 1 in 500. In this case the amboceptor alone was effective in the same strength and the "606 alone" was effective in 1 in 100.

From this it follows that *in vitro* the trypanocidal action of the amboceptor is not added to the trypanocidal action of the arsenic compound. If that were the case the complex should exceed either of the components in activity. It must therefore be concluded that amboceptor and "606" do not act upon the trypanosomes in the same manner. In EHRLICH's terminology, the arsenic receptor is different from the receptor acted upon by the trypanolytic antibody.

(2) "606 alone" serum exercises markedly different effects *in vitro* and *in vivo*. While in the majority of cases this serum when injected almost at the same time as the trypanosomes, but at a different part of the body, did not prevent the evolution of the infection nor a fatal termination, *in vitro* it caused the destruction of the trypanosomes.

This difference of action can only be explained on the supposition that new properties are acquired by the "606" when it is subjected to the action of the cells and fluid of the body. It is probable that its diffusibility is altered and also its affinities for the tissues.

It is quite another matter when the arsenic compound is present in the serum in association with amboceptor. The results obtained *in vitro* and *in vivo* force one to the conclusion that in the animal body the action of one of the components of the complex assists the action of the other. Further, it appears that the two trypanocidal agents acting upon the two sets of receptors should render relapses of arsenic and antibody-resistant strains less likely and thus lead to complete sterilisation. Arsenic-fast individuals will be subjected to the toxic influence of the antibody trypanocides and vice versa. These facts may have important practical consequences.

The authors have the idea that in sera containing the "606" and amboceptor these two substances are associated with the same protein molecule, but they have not been able to get experimental results plainly indicating this. All that they have been able to shew is that the arsenical compound and the amboceptor cannot be separated by dialysis through collodion sacs, and further, that precipitation of the albumins and globulins does not lead to distinct separation. Though this suggests that the attachment may be to the same protein

molecule, a simple explanation, it is admitted that the experimental facts could be explained in another manner, such as simple absorption and physical inclusion of the arsenic compound by the precipitates.

Whatever may be the real explanation, the fact remains that the results obtained indicate a line which may have valuable practical results.

- (333) JACK (R. W.). **Tsetse Fly and Big Game in Southern Rhodesia.**
—*Bull. Entomol. Research.* 1914. Sept. Vol. 5. No. 2. pp. 97–110.
With 2 maps.

This paper contains an account of the investigations of the author regarding the connection between *G. morsitans* and big game. During the last four and a half years he has visited the great majority of the fly-belts in the territory, and the main belts have been visited repeatedly. The broad result of the observations is that game is more or less abundant all the year round in the fly-infested country, and that in no instances is the larger animal life absent, even during a portion of the year. The evidence collected is considered to be strongly in favour of a vital association between the prevalence of big game and the continuance and increase of the fly.

The evidence in favour of the necessity of big game to the tsetse in Southern Rhodesia and the adjacent territory may be summed up under four heads :—

1. Tsetse retired before the advance of civilisation in the Transvaal, the only known modification of conditions being the destruction of game.
2. Tsetse disappeared from large tracts of country immediately after the rinderpest epizootic of 1896.
3. Tsetse has increased and spread since the rinderpest only in those parts of Southern Rhodesia where big game has increased.
4. Tsetse has greatly decreased of late years in the Hartley District in those parts where big game has been most effectively destroyed or driven away.

LEISHMANIASIS.

- (334) ROW (R.). **Experimental Leishmaniasis in the Monkey and the Mouse induced by the Parasites in Culture.**—*Indian Jl. Med. Research.* 1914. Apr. Vol. 1. No. 4. pp. 617–621. With 2 coloured plates.

This paper contains an account of experiments in which susceptible animals were infected with cultures of *L. donovani*, and of *L. tropica*. The principal object in recording the experiments is that they afford an easy means of testing the infectivity of this or that intervening host which may be brought forward from time to time as the agency responsible for the transmission of the disease to man.

A.—Kala azar culture experiments.—

The success or failure of experimental infection with cultures appears

to depend on the susceptibility of the animal used, the age and morphological phase of the parasite in culture, and the method of infection.

The smaller race of *Macacus sinicus* found in the neighbourhood of Madras is more susceptible than the larger race from the Bombay district, and this animal appears to be the subject *par excellence* for experimental purposes.

The author has not had any successes in attempting to infect dogs, although LAVERAN working with the same strain has infected three out of eight.

The author's observations have convinced him that positive results are only obtained when the cultures contain large numbers of small rounded bodies resembling the original parasites, or small coccus-like bodies which look like "fused condensed nuclei of the parasites enveloped in a mere suspicion of a zone of body protoplasm."

These results differ from those obtained by NICOLLE, MANCEAUX, LAVERAN, and GONDER, who have only obtained positive results by using young active flagellates.

Intraperitoneal inoculations if successful always produce a generalised infection, while subcutaneous inoculation leads to a localised lesion not followed by generalisation, or generalisation preceded either by a very slight localised lesion or none at all.

B. Oriental Sore culture experiments.—

The author states that, while he has been able to infect monkeys experimentally with *Leishmania tropica* from cases of Cambay sore, he has never been able to infect them with cultures of the parasite. NICOLLE and others have recorded successful inoculation of the monkey with the production of a local lesion, and GONDER has recorded infection of the spleen and liver in an inoculated mouse, with localisation at a later period.

The author has subjected a mouse to the following inoculations :—

| | | | | |
|---------------|-------------------|---------|--------------------|----------------------|
| 1st injection | intraperitoneally | 9-4-13, | 0.5 cc., | 7 weeks old culture. |
| 2nd | " | " | 10-4-13, 0.25 cc., | 4 " " |
| 3rd | " | " | 11-4-13, 0.25 cc., | 4 " " |
| 4th | " | " | 11-6-13, 0.25 cc., | 6 " " |

All the cultures were rich in rounded forms and contained hardly any motile forms.

The animal remained alive and apparently healthy until it was killed 9 months and 13 days later.

At the post-mortem the spleen was six to eight times the normal size and the liver was also somewhat enlarged. The peritoneum and the other organs appeared normal. Smears from the liver, spleen, and bone marrow showed an exceedingly rich infection, and cultures yielded pure flagellates. In smears made from the heart blood large coccus-like bodies were found in the red corpuscles. These bodies were not cocci, as they would not grow upon agar.

These facts show that, like *Leishmania donovani*, *Leishmania tropica* in culture produces a generalised infection in the mouse, while kala azar cultures administered subcutaneously produce a local lesion, or a generalised infection, or both.

The question arises whether the parasites are identical and the author expresses the hope that further experiment will throw light upon the subject.

- (335) KORKE (V. T.). A Note on the Production of Localised Lesions by *Leishmania donovani* in *Macacus sinicus*.—*Indian Jl. Med. Research*. 1914. Apr. Vol. 1. No. 4. pp. 622-625.

This short paper contains brief details of a number of experiments carried out to check Row's results regarding the production of localised lesions in monkeys with *Leishmania donovani*.

The conclusions arrived at are as follows :—

"1. In *Macacus sinicus*, *Leishmania donovani* is capable of producing purely localised lesions which take the form of nodules.

"2. Injecting infective material into pockets of skin on the foreheads of monkeys is an effectual and easy way of producing such lesions. Such a procedure may at times give rise to a systemic or generalised infection.

"3. The incubation period of these nodules varies between 52 and 71 days.

"4. The *Leishmania* contained in such nodules are capable of giving rise to similar lesions in other monkeys. They appear in typical form as well as mononuclear round bodies. Occasionally parasites in these nodules are very small though typical in other morphological respects."

- (336) MACKIE (F. P.). Kala azar in Nowgong (Assam).—*Indian Jl. Med. Research*. 1914. Apr. Vol. 1. No. 4. pp. 626-662. With 6 maps and 6 charts.

This paper is a report of the work done during the period February to September, 1912. It is interesting to note that no connection was traced between the disease in man and its occurrence in the dog or any other species of animal.

- (337) PATTON (W. S.). The Behaviour of the Parasite of Indian Kala Azar in the Dog Flea, *Ctenocephalus felis*, Bouché, with some Remarks on Canine Kala Azar and its Relation to the Human Disease.—*Indian Jl. Med. Research*. 1914. July. Vol. 2. No. 1. pp. 399-403.

The material examined was obtained from about 200 fleas which were fed upon a dog infected with the human parasite during the four days preceding its death. Four days before death about 500 parasites could be found in every smear of peripheral blood, and two days before death there were about 1,000 parasites in every such film. In Madras the dog flea is *Ctenocephalus felis*. *Ctenocephalus canis* is found on the jackal only.

The fleas were dissected at varying intervals after the meal of infected blood, and it was found to be exceedingly difficult to find parasites eight hours after the meal, and none were found in the fleas dissected at later intervals.

The conclusions drawn are as follows :—

"1. The parasite of Indian kala azar does not develop in the dog flea, *Ctenocephalus felis*, but degenerates and disappears in eight hours. This together with the fact that the dog has not been found infected with kala azar, or perhaps, to be exact herpetomoniasis, in India, strongly supports the view that human kala azar is not of canine origin. The human flea, *Pulex irritans*, has not been found in Madras.

"2. Assuming that the parasites of Indian and Mediterranean kala azar are identical, it is difficult to understand why the one does not develop in the flea, while the other does.

"3. The fact that the dog may be infected with *Herpetomonas clenoccephali* suggests that the so-called canine kala azar may have nothing to do with the human disease, and that its association with it is a coincidence. There is at present no proof that either *Ctenocephalus canis* or *Pulex irritans* transmits the parasite of human kala azar from dog to man and man to dog, and the hypothesis advanced by NICOLLE and so vigorously supported by BASILE and others appears to be based on very slender evidence."

- (338) SERGENT (Edm.), SERGENT (Et.), LEMAIRE (G.) & SENEVET (G.)
Insecte Transmetteur et Réservoir de Virus du Clou de Biskra.
Hypothèse et Expériences Préliminaires. [Insect Transmitting
 Agent and Reservoir of the Virus of Oriental Boil at Biskra.
 Hypothesis and Preliminary Experiments.]—*Bull. Soc. Path.*
Exot. 1914. July. Vol. 7. No. 7. pp. 577-579.

Epidemiological observation does not allow of any view regarding the transmission of oriental sore at Biskra other than that it is transmitted by a biting insect, and, in view of the fact that the exposed portions of the body are the predilection seats of the boils, the insect is in all probability winged.

In 1904, the authors incriminated *Phlebotomus* as being the transmitting agent, and since that date other authors have published papers agreeing with this view.

Since oriental boil is a localised affection (GONDER's view that it is a generalised disease being unproved), it is scarcely possible to hold that the human subject is the reservoir whence the insects derive the virus. A reservoir must therefore be sought among the animals which are bitten by *Phlebotomus*.

Phlebotomus of the group *P. papatasi* bite the human subject for preference or perhaps exclusively. But those of the group *P. minutus* normally bite reptiles.

Two species are encountered at Biskra: *P. papatasi* and *P. minutus*, var. *africanus*. The Geckonidae, which have been described by HOWLETT in India as the natural host of *P. minutus*, are very commonly found in houses in Biskra, the species being *Tarentola mauritanica*.

The authors have carried out investigations with the idea of testing the hypotheses that *P. minutus* may be the transmitting agent and the tarentola the reservoir of the virus.

The seasonal occurrence of the disease may be associated with the fact mentioned by HOWLETT that *P. minutus* only bites man during the hot season, and during the cold weather lives on the hibernating geckos.

The authors have verified the statement that *P. minutus* feeds on the tarentola at Biskra, and they have also found that it bites the human subject.

At the moment their investigations permit them to say that in nearly 16 per cent. of cases cultures made from the organs of the tarentola yield pure cultures of a leptomonas resembling the leptomonas of cultures of the organism of oriental boil. Interest attaches to this point because all the cultures of leptomonas so far obtained from the organs of vertebrates have been derived from leishmania.

- (339) ARCHIVES DE L'INSTITUT PASTEUR DE TUNIS.—1914. Aug. 1. Vol. 9. No. 1. pp. 30–38.—*Chronique de Kala Azar en Tunisie*. [Statistics of Kala azar in Tunis].

Since the publication of the last statistics eight fresh cases of leishmaniasis in the human subject have been discovered in Tunis and one case in Algeria. The majority of the cases have been found in boys under three years of age.

During the year 1913, 109 dogs which presented symptoms suggestive of infection were examined, and of these six were found to be infected.

Five strains of parasites are under cultivation. Two of these are from human sources and have reached the 100th and 56th generation respectively, two are from canine cases and have passed through 61 and 58 subcultures, and the remaining strain is from a case of oriental sore which is in the 119th generation. The reinoculations are made twice monthly on blood agar. The last of the strains was isolated in 1909, and during the period that it has been under artificial cultivation it has not undergone any change of morphology.

It has been found very difficult to maintain the canine strain by passage through dogs, and it is a curious fact that the canine strain of the parasite is less infective for the dog than the human strain.

SPIROCHAETOSIS.

- (340) WOLBACH (S. B.). *The Distribution and Morphology of Spirochaeta duttoni and Spirochaeta kochi in experimentally infected Ticks (Ornithodoros moubata)*.—*Jl. Med. Research*. 1914. March. Vol. 30. No. 1. pp. 37–48. With 3 plates comprising 18 figures.

The investigations described in this paper were rendered possible by the elaboration of a special method of staining with Giemsa.

The full details of the method are given and the following are the principal points:—

The tissues are fixed in sublimate alcohol and are passed through into paraffin. Sections should be 2 microns in thickness. After complete removal of the mercury by iodine, and of the iodine by hyposulphite of soda, the sections are stained with the following dilution of Giemsa: sixty drops of stain, 100 cc. of distilled water, 10 cc. of methyl alcohol and 2 drops of .5 per cent. sodium carbonate. The stain is replaced twice during the first hour and then left acting for 20 hours.

After staining the sections are differentiated in two changes of 100 cc. of acetone containing 15 gm. of colophonium. The process takes from 15 to 30 seconds. The sections are then passed through acetone xylol, pure xylol, and mounted in cedar oil. Uniform results have been obtained.

As a result of the examination of preparations made from experimentally infected ticks treated in this way the following conclusions are drawn :—

“ 1. The modified Giemsa stain employed is a reliable method of demonstrating spirochaetes in the tissues of ticks.

“ 2. The two strains of spirochaetes studied, *Spirochaeta duttoni* and *Spirochaeta kochi*, migrate through all the organs and tissues of the ticks.

“ 3. Multiplication of spirochaetes does not take place in the epithelial cells of the tick, but does occur in the tissues of the connective tissue group.

“ 4. The minute granules and comma bodies found in epithelial cells, and probably those in other tissues, are not stages in the development of the spirochaetes.

“ 5. Large granules, coiled and encysted forms derived from spirochaetes, occur in various connective tissue structures, and may possibly represent resting or multiplication stages.

“ 6. By virtue of the power to invade tissues, the spirochaetes studied may be transmitted in any secretory or excretory product of the ticks.”

(341) TODD (J. L.) & WOLBACH (S. B.). Concerning the Filterability of *Spirochaeta duttoni*.—*Jl. Med. Research*. 1914. March. Vol. 30. No. 1. pp. 27-36.

Following the technique employed by NOVY and KNAPP, BREINL and KINGHORN, the authors have repeated the filtration experiments of these authors and have arrived at the following conclusions :—

“ 1. *Spirochaeta duttoni* in an infective form can be forced through a Berkefeld filter by pressures of over fifty pounds to the square inch.

“ 2. *Spirochaeta duttoni* cannot be filtered through a Berkefeld filter, in an infective form by atmospheric pressure.”

MALARIAL AND OTHER BLOOD PARASITES.

(342) PHISALIX (Mme). Sur une Hémogrégarine Nouvelle, Parasite de *Boodon fuliginosus* Boë, et ses Formes de Multiplication Endogène. [A New Haemogregarine parasitic in *Boodon fuliginosus*, and its Endogenous Multiplication Forms.]—*Bull. Soc. Path. Exot.* 1914. July. Vol. 7. No. 7. pp. 575-577.

The snake in whose blood the parasites described in this paper were found was sent from the Soudan. It died in an extremely anaemic condition a month after its arrival.

Apart from the blood parasites the following were also found :—

On the skin *Ophionyssus natricis* and *Aponema laeve* were present, the former being in large numbers.

Numerous slender nematodes, the species of which has not yet been determined, were found in the oesophagus and in the connective tissue surrounding the viscera.

Examination of the blood showed that there was profound anaemia and that the majority of the remaining blood corpuscles had lost the greater part of their haemoglobin.

The haemogregarine was present in the peripheral and heart blood in very considerable numbers, but no multiplication forms were found in the latter. They were, however, found in the capillaries of the lungs, liver, kidneys, spleen and pancreas.

Two endoglobular forms were recognisable.

(a.) Vermicules measuring 14 to 15 microns in length, and 2 to 3 in width. These were slightly curved at the extremities, one of which tapered slightly. They were found applied to the nucleus of the cells, and possessed a thin unstained capsule.

The nucleus formed an elongated mass which was sometimes bilobed. It was placed close to the convex border of the parasite towards the thicker end. It was homogeneous and stained deeply. In some instances two parasites were found in a single corpuscle. The corpuscles did not appear to have undergone any alteration save an increase in size due to the presence of the parasites.

(b.) In a minority of cases parasites of the same length, but of three or four times the thickness, were found. In these the nucleus was rounded or annular, and composed of filaments of chromatin. These stained with moderate intensity, and in some cases there were granules scattered through the protoplasm which did not take the stain.

Free forms were represented by both of these types. In the majority of cases they were only set free by a gradual disintegration of the blood corpuscle containing them. Subsequently the parasites themselves tended to degenerate, first becoming swollen and then disintegrating. Those parasites which had previously been slender could be recognised by the slight curving of their extremities.

Cysts containing macromerozoites were encountered in small numbers in the lungs, liver, spleen, and kidneys. They were regularly ellipsoid in shape and contained 2 or 4, and rarely 6, merozoites. The smallest of these measured about 17 by 12 microns, and the largest about 30 by 20. The nuclei stained intensely, and the protoplasm very distinctly. The capsule did not stain.

Cysts containing micromerozoites were found in large numbers in the lungs, liver, and pancreas, but not in the spleen and kidneys. The youngest, corresponding to the first stages of division of the nucleus, contained 2 or 4 nuclei embedded in a granular mass. They measured about 25 by 18 microns. Those in the most advanced stage of development were packed with little ovoid nuclei measuring about 1 micron. These stained intensely. Cysts in this stage measured 30 by 25 microns. The envelope was very thin, and around a great number of them, but not all, there could be seen a retraction zone in which the parasite occupied an eccentric position.

None of the cysts were found to be in the process of rupture, and no forms of exogenous multiplication were seen.

The name *Haemogregarina boodoni* is suggested for the parasite.

- (343) ACTON (H. W.) & KNOWLES (R.). Studies on the Halteridium Parasite of the Pigeon, *Haemoproteus columbae*, Celli and San Felice. *Indian Jl. Med. Research*. 1914. Apr. Vol. 1. No. 4. pp. 663-690. With 5 coloured plates and 5 tables.

In a paper published in 1913 on the "Diagnosis of Latent Malaria," the authors suggested that the terms relapse and recrudescence should

be differentiated from each other. Relapse was defined as a biological phenomenon involving the re-formation of asexual forms from the and persistent gametes; whilst they regarded recrudescence as an resistant exacerbation of the asexual cycle.

As material from human sources was not available to afford proof of these views, experiments had to be carried out and investigations made regarding malarial infections in animals.

After having examined a number of monkeys for malarial infection without success and having found that sparrows infected with *Proteosoma* did not live in captivity for more than two or three days, the authors fell back upon *Halteridium columbae*, which was the only parasite analogous to *Plasmodium malariae* available.

From their studies in connection with the life-cycle of the parasite in the pigeon the authors have come to the conclusion that the description given by BEAUREPAIRE-ARAGAO is misleading, and that that given by NEGRI is correct for the mature schizont stage only. The figures given in DOFLEIN after LABBÉ are inaccurate, and in their opinion, represent the life-cycle of the *Proteosoma* parasite.

The details of the asexual cycle are said to be very puzzling, but the authors believe that their description of it is accurate in its essential details.

The asexual cycle.—The merozoites are small, ovoid or irregular bodies measuring about 2 microns in diameter, and consist of a small granule of chromatin with fairly darkly staining cytoplasm. They invade red corpuscles in the capillaries of the lungs and develop into trophozoites. These are oval in shape and measure from 3 to 5 microns in diameter. The chromatin is ovoid in shape and is situated near the narrower pole of the parasite. The cytoplasm stains faintly and contains very few pigment granules. They mature into schizonts.

The schizont rapidly increases in size and eventually obstructs the capillary in which it has been arrested, and the chromatin undergoes a process of fragmentation by binary fission. No pigment is observed in these stages, and in the centre of the schizont a large faintly stained mass of chromatin is seen.

The merozoites appear to escape by the bursting of the spore sac, and they carry on the asexual cycle by infecting fresh red corpuscles.

The asexual cycle appear to occur nowhere save in the lungs.

The merozoites give rise to gametes which require 6 to 10 days to come to maturity. These gametes are specially resistant forms. The author believe that only gametes are found in the peripheral blood because conditions there are not favourable for schizont formation, and that therefore young merozoites which are carried into the general circulation become gametes.

The macrogametocyte.—In this stage the parasite is elongated and it grows until it reaches the typical shape. The nucleus is centrally placed and stains a bright crimson colour. There are small light brown pigment granules scattered irregularly through the parasite.

The microgametocyte.—The young parasite is oval in shape, with a diffuse irregular orange staining chromatin mass. The cytoplasm is hyaline and stains of a faint purple colour. There may be one or two pigment granules, which are large. As the parasite increases in size the pigment granules become more numerous until 4 to 10 may be seen at each end of the parasite.

In opposition to the views expressed by Cragg* in 1907, the authors state that (1) they have never seen intracorpuseular conjugation of trophozoites in the Halteridium; (2) The resting or "zygote" stage described by him has never been observed, but they consider the adult gamete as the resting stage and resistant form of the parasite, which alone is able to withstand injurious influences; (3) They regard the gamete as the originator of relapses.

The multiplication of the parasite in the capillaries of the lungs leads to the production of macroscopic changes in the appearance of these organs at the post-mortem, and they believe that in this fact there possibly lies an explanation of the frequency of pneumonia among malarial cachectics.

The authors believe that multiple infection of a single corpusele is a matter of chance, and that though cytoplasmic union may occur each parasite maintains its individuality.

During the winter 1912-13, the authors spent several months trying to confirm the asexual cycle as described by BEAUREPAIRE-ARAGAO, but entirely failed to do so. The reason for this was that they were working with pigeons which had been kept for several months under good conditions, and which had only mature gametes in their blood.

Early in 1913, a fresh batch of birds was obtained and in some of these young gametes were found in the peripheral blood. The authors were then able to work out the asexual cycle in the lung as described in this paper.

This observation was so repeatedly confirmed that they are able to rely upon the discovery of young gametes in the blood as an index to the presence of mature schizonts in the lungs. Where only mature gametes are present in the blood for a number of days it is found that asexual development in the lung has ceased.

A number of experiments were carried out in which blood was withdrawn from infected birds shewing either immature or mature gametes in their blood, the number of parasites being estimated from day to day. If there was an increase in the number after the taking of the blood it was an indication that asexual multiplication in the lung had not ceased and vice versa, because when only mature gametes are present in the blood there cannot be any production of new generations in the lungs by schizogony.

Though they have not been able to ascertain any maximum period for which gametes may persist in the blood, they think that it may be for some months or perhaps longer. By virtue of their great resistance and prolonged life the gametes enable the parasite to live through the winter months and bridge the interval from one fly season to another.

The authors made a number of observations upon the sex differentiation of the gametes present, and in 6 out of 10 pigeons examined the counts seemed to shew a typical Mendelian distribution; the female being dominant and the male the recessive.

In this disease the evidence points to the spleen as being the seat of destruction of the gametes.

In endeavouring to obtain evidence that female gametes can and do give rise to asexual forms, which would explain the occurrence of relapses after long intervals when fresh infection can be excluded,

* CRAIG is obviously meant.

the authors have used BASS's method of cultivating the halteridium of the pigeon.

Examination of the blood of a very large number of pigeons convinced the authors that it is in very exceptional cases only (they found one instance) that trophozoites can be found in the peripheral blood; they therefore made cultures from blood in which only gametes could be found on a number of successive days.

The blood was drawn from the marginal vein of the wing into an equal quantity of .85 per cent. salt solution containing 1.5 per cent. of sodium citrate. This mixture was placed in sterile tubes containing 50 per cent. dextrose (Merck) solution in the proportion of 5 cc. of the mixture to 1-10 cc. of dextrose. The tubes were incubated in an atmosphere of hydrogen at 40°-42°C. and examined every two hours. Preparations were made from the layer of corpuscles immediately below the serum.

It appeared to be important that the blood should be allowed to cool in the syringe for a few minutes "to give the necessary stimulus to the process."

Appearances were found which indicated that asexual parasites did actually develop from the female gametes, and that these gave rise to merozoites.

In one instance an attempt was made to cultivate young gametes found in the blood to adult ones, but without success. The young gametes did not, however, assume a trophozoite-like form, thus shewing that the trophozoite-like forms which were observed in other cultures did not develop from young gametes.

The trophozoite forms appearing in the cultures were not present in the blood when taken from the bird, because control preparations shewed none, and, further, because ten out of twenty-one cultures shewed trophozoite forms, while trophozoites were only once found in the blood out of more than 200 pigeons examined.

The male gametes could not have taken part in the process as the conditions necessary for the formation of microgametes were not present and all the male parasites were observed to degenerate very rapidly in cultures and to be ingested by phagocytes.

The authors consider the change of temperature and environment form the stimulus to the female parasites to become parasites acting as trophozoites, and in this they find the explanation of the relapses occurring in malaria in the human subject, as such relapses always occur when circumstances become unfavourable to the host, and when the blood environment of the parasite (as for example when vaccines are introduced) is altered.

- (344) LEGER (A.) & LEGER (M.). *Sur un Plasmodium de la Roussette du Haut-Sénégal et Niger*. [A Plasmodium of the "Flying-Fox" of the Upper Senegal and the Niger.]-*Compt. Rend. Soc. Biol.* 1914. July 24. Vol. 77. No. 26. pp. 399-401.

The authors have found parasites which they have identified with the parasite, *Pteropus gouldi*, described by BREINL in the Report of the Australian Institute of Tropical Medicine, 1911, as occurring in the Australian "flying-fox."

- (345) MACKIE (F. P.). Note on the Parasite of Bat Malaria.—*Indian Jl. Med. Research*. 1914. July. Vol. 2. No. 1. pp. 375-376. With 1 coloured plate.

The parasite described in this note was found in the blood of a flying fox (*Pteropus edwardsii*). The animal was kept under observation for several weeks without showing any symptoms of illness. The parasite closely resembled the malarial parasite of the African monkeys. The majority of those present in the blood were in the adult stage, and gametocytes were numerous. The addition of saline solution to the blood enabled the process of flagellation to be observed. The microgametes resembled those of the parasite of human malaria.

The infected animal was kept in a cage with others, but the infection did not spread.

In view of the fact that since the infected animal was found 56 others have been examined without success, it would appear that the parasite is somewhat rare. The name, *Plasmodium pteropi* (n. sp.), is suggested for the parasite.

HELMINTHS.

- (346) BOYNTON (W. H.). Kidney-Worm Infestation of Swine in the Philippine Islands with Special Reference to the Pathological Changes.—*Philippine Jl. Science*. Sec. B. Trop. Med. 1914. June. Vol. 9. No. 3. pp. 269-289. With 3 plates.

The condition described in this paper is far more commonly met with in sub-tropical and tropical climates than in colder countries. It has been observed in the United States, especially in the southern part, South America, Australia, Asia, and in the Philippine Islands.

The cause of the condition is *Stephanurus dentatus*, also known as *Sclerostoma pinguicola*.

Examination of 2,000 pigs slaughtered at Manila showed that nearly 50 per cent. of the animals were affected.

The symptoms shown are: stiffness, especially of the hind limbs, the lameness and loss of power increasing progressively. There may be almost complete loss of power in the hind limbs a few days before death. The appetite is generally maintained until a few days before death.

The parasite has been encountered in a number of different situations. It occurs free upon, and embedded in, the fat surrounding the kidneys, and also within the kidney itself. It occurs free in the peritoneal cavity, in the liver, portal and mesenteric glands, and especially in great numbers in the connective tissue along the back. It has been encountered in the pleural cavity and embedded in the pleura.

It is said that in infested carcasses putrefaction occurs very rapidly.

A detailed description is given of the changes produced in the organs resulting from invasion by the worm.

- (347) LANE (C.). **Bursate Nematodes from the Indian Elephant.**—*Indian Jl. Med. Research.* 1914. July. Vol. 2. No. 1. pp. 380–398. With 12 plates.

The author of this paper has had the opportunity of examining material from six elephants and has found eight species of bursate worms, of which six are new and fall into six genera.

As full a description as possible is given of each, and the paper is illustrated with numerous plates.

FILTERABLE VIRUSES.

(RINDERPEST AND INFECTIOUS ANAEMIA OF HORSES).

- (348) BOYNTON (W. H.). **A Preliminary Report of Experiments on the Cultivation of the Virus of Rinderpest in Vitro.**—*Philippine Jl. Science.* Sec. B. Trop. Med. 1914. Feb. Vol. 9. No. 1. pp. 39–44.

In preliminary experiments it was found that sterile blood drawn from infected animals maintained its virulence better under anaerobic than under aerobic conditions, and also that the virulence was maintained for a longer time when the blood was withdrawn within a day or two of the first rise of temperature than at later stages.

Experiments showed that neither red corpuscles, white corpuscles, nor blood platelets transmitted the disease, nor did they confer any immunity.

Investigations were then carried out to ascertain the smallest quantity of blood that would set up the disease. By the use of very fine calibrated pipettes it was found that 1/2970 cc. of virulent blood was capable of setting up the disease, but that 1/9060 failed. These data were useful in the cultivation experiments for comparison with the dilution of the original blood brought about by successive transfers in culture media.

In a number of media that were first tried it was found that the virus died in from 36 to 68 hours, but positive results were subsequently obtained with a modified form of the medium described by NENCKI, SIEBER, and WIJNKEWITCH, and with the medium used by BASS and JOHNS.

Nencki's medium, which consisted of 900 cc. of water containing 100 grammes of peptone and 20 grammes of sodium chloride, yielded negative results.

This medium was modified by the addition of 33½ per cent. solution of glucose to the peptone water in the proportion of 0.1 cc. to 10 cc. Immediately before use normal sterile defibrinated blood was added to the tubes in the proportion of 1 cc. to each tube of 10 cc. These tubes were then inoculated with 0.5 or 1 cc. of virulent blood, anaerobic conditions being produced by putting a layer of sterile paraffin oil on the surface of the medium. The tubes were incubated at 40° C. Subinoculations were made every three or four days, using sterile pipettes and transferring 0.3 to 0.5 cc.

The medium giving the best results was a modification of that used by BASS and JOHNS for the cultivation of malarial parasites. Glucose solution was added to sterile defibrinated blood taken from a susceptible animal in the proportion of 0.1 cc. to 10 cc. This medium was inoculated and then covered with sterile paraffin and incubated at 40° C.

The sixth transfer covering a period of 21 days in the blood glucose medium was found to be virulent in one instance, and in another the sixth transfer at 19 days was found to be virulent. In the salt peptone mixture inoculation succeeded with the fourth transfer at 12 and 13 days.

To ascertain whether the successful inoculations were due to original virus, calculations were made to ascertain the dilution, and in one case the virulent dose of the sixth transfer was calculated to contain 1/2,832,921 cc. of original virus. This suggests that there was an actual multiplication.

Nothing could be discovered regarding the morphology of the virus.

One animal which recovered from the disease induced by a fourth transfer is being hyperimmunised with cultures with the object of testing its serum. Experiments are also being carried out to test the possibility of devising a combined method of immunising with cultures and serum.

(349) BOYNTON (W. H.). Experiments on the Cultivation of Rinderpest Virus as described by Baldrey.—*The Philippine Jl. Science*. Sec. B. Trop. Med. 1914. June. Vol. 9. No. 3. pp. 259-268.

BALDREY's paper, which was published in the *Journal of Tropical Veterinary Science*, Vol. 6, No. 1, 1911, contained an account of some experiments in which rinderpest blood was mixed with broth and incubated for 24 hours at body temperature, the liquid thus obtained being used for the hyperimmunisation of animals.

BALDREY arrived at the conclusion that an active toxin was produced in the broth, and that this toxin killed off the virus. It was found that it was best to inject the material intravenously, as if given subcutaneously it produced very severe inflammatory lesions.

The author of the present paper has repeated BALDREY's experiments, and his conclusions, excluding references to particular experiments, are as follows:—

"1. In the case of two animals which died in less time than the incubation period of rinderpest, after injection of the Martin's broth culture, the autopsy findings of the tissues indicated death from a bacterial infection and not from rinderpest. All evidence points to the conclusion that the Martin's broth employed in these two cases was contaminated by bacteria prior to injection in the animals. The results are attributed to poor aseptic technique, and greater care in subsequent inoculations, where no such toxæmias were induced in the injected animals, support the conclusion.

"The symptoms, lesions, and other circumstances stated by BALDREY resemble the results obtained in the two animals in question, and there is justification for belief that his results were due to the same cause.

"2. In all the other animals injected with mixtures of blood and culture medium after incubation, no immediate ill effect followed in either susceptible or immune animals.

"3. With the exception of the animals noted in these experiments, all those injected with the so-called twenty-four- and forty-eight-hour cultures

of rinderpest in neutral or alkaline Martin's broth contracted rinderpest after the usual incubation period and died. These observations do not support BALDREY'S belief that there occurs a rapid formation of rinderpest toxin in the broth during the twenty-four hours with the resulting death of the virus. The experiments have included tests of Martin's broth after incubation as long as seventy-two hours.

"Rinderpest virus does die in Martin's broth culture after incubation for seventy-two hours, but there is no evidence that rinderpest toxin was formed, much less that rinderpest toxin caused the death of the virus.

"4. The experiments reveal the fact that rinderpest virus will survive in neutral or alkaline Martin's broth at 37°C. for at least forty-eight hours, but not for seventy-two hours. Two cases were tested at twenty-four hours, 2 at forty-eight hours, and 3 at seventy-two hours.

"5. Rinderpest virus kept in acid Martin's broth or in 5 per cent. potassium citrate solution did not survive after forty-eight hours at 37°C."

(350) BOYNTON (W. H.). *An Atypical Case of Rinderpest in a Carabao.*—*Philippine Jl. Science.* Sec. B. Trop. Med. 1914. Feb. Vol. 9. No. 1. pp. 45-47. With 1 chart.

The facts regarding the peculiar case of rinderpest described in this article are summarised in the conclusions and are as follows:—

"1. From the facts of this case, the evidence is conclusive that an animal may experience a fatal attack of rinderpest without the occurrence of a rise in temperature.

"2. The blood of the carabao was shown to be infected within forty-eight hours after it was originally injected with virulent blood.

"3. It was shown that the blood was virulent on the eleventh day when injected into a susceptible animal, yet exposure to the same animal from which blood was drawn did not cause rinderpest in the exposed susceptible animal.

"4. With regard to the three animals which failed to contract rinderpest by exposure, the question is raised as to whether rinderpest spreads by contact readily in the later stages of the disease or whether the disease must necessarily be accompanied by a rise of temperature before it can spread by contact."

(351) WARD (A. R.), WOOD (W. F.) & BOYNTON (W. H.). *Experiments upon the Transmission of Rinderpest.*—*Philippine Jl. Science.* Sec. B. Trop. Med. 1914. Feb. Vol. 9. No. 1. pp. 49-78. With 2 plates and 6 charts.

The experiments described in this paper were designed to supply answers to the questions as to the period for which the virus can retain its vitality outside the body under varying natural conditions, and also the period during the course of the disease in which the virus is disseminated by sick animals.

The following conclusions were arrived at:—

"1. Rinderpest virus was not shown to have survived beyond twenty-four hours in corrals bare of vegetation but containing water. The conditions under which the tests were made included all seasons of the year, with accompanying variation in sunlight, rain, and condition of the soil. The amount of shade varied widely.

"2. Animals in such corrals became infected with rinderpest within half an hour, twelve hours, and seventeen and one-half, respectively, after the removal of the sick.

"3. Animals infected with rinderpest were shown to be capable of transmitting the disease to susceptible animals by close contact only during the febrile period of the disease, and most certainly during the period when the temperature was declining. The disease was not contracted

by susceptible animals when exposed to sick animals during the convalescent stage when the temperature was nearly normal.

"4. Blood of animals infected with rinderpest was shown in two cases to be infected during the height of the febrile period.

"5. The virus in urine, diluted with water and sprinkled on grass, was demonstrated to survive for thirty-six hours in some instances, but not always, and not for a longer period of time.

"6. Faeces mixed with water and sprinkled on grass infected an animal twenty-four hours later.

"7. Faeces and urine diluted with water and kept in a vessel in the shade remained infective for susceptible animals for thirty-six hours, but no longer.

"8. No evidence was secured to show that recovered cases transmit the disease.

"9. The foregoing facts indicate that the virus of rinderpest perishes soon after being discharged by the infected animal.

"10. Nothing in the foregoing experiments indicates that rinderpest virus is harboured for long periods upon the soil of contaminated areas."

(352) **Report on the Results obtained by the Special Committee for the Investigation of Infectious Anaemia among Horses.**—The Horse Administration Bureau. Tokyo. 1914. pp. 1-59. With 9 plates, 3 charts and 1 map.

The Special Committee, whose report this is, was appointed by the Japanese Government for the investigation of pernicious anaemia in horses in 1909. The personnel of the committee included representatives of the Horse Administration Bureau, the Imperial University at Tokyo, the Department of Agriculture, and the Army Veterinary Service.

Although exact information was not obtainable, it appears to be probably that infectious anaemia among horses was unknown in Japan prior to 1895.

The disease seems to be confined to Hokkaido, the northernmost island, and the north eastern part of Hondo, the main island. The southern portion of Japan is not invaded, because this portion of the country contains no pasture land, and it is among horses at pasture that the disease spreads.

Exact information is also lacking regarding the annual losses from the disease. In the first year of any outbreak the number of cases is small. This is followed by a progressive increase during the following years until the disease may be so severe that the majority of the horses in the district die. The disease then slowly dies down again. Once infected a district remains infected for a number of years, cases having been recorded in some districts for more than ten consecutive years after the initial outbreak.

In 1907, it was recognised that a number of deaths among the horses belonging to the Remount Dépôt, which had been attributed to other causes in previous years, were in reality due to infectious anaemia. The maximum losses occurred in 1910 when a total of 500 horses died or were killed. Since that date the number of cases has decreased as a result of the preventive measures put into force. Since the virus is generally introduced by two-year-olds which have every appearance of being healthy at the time of purchase, but which afterwards prove to be infected, complete success in stamping out the disease among the remount animals has not yet been achieved, and fairly heavy losses still occur.

It is while horses are at pasture that the majority contract the infection, animals very rarely becoming infected in stables. Unless death occurs soon after infection, the disease takes a chronic course. During the first few months after infection repeated febrile attacks occur, but in cases in which the animal survives these gradually decrease in severity and finally cease.

The virus was found to be capable of passing through a Berkefeld filter, and all attempts to cultivate it outside the body failed. Experimental inoculations shewed that the virus is present in the blood and all the organs, and also in the urine and milk, but attempts to prove its presence in the faeces and sweat of infected animals failed.

That the virus is protozal rather than bacterial in nature is indicated by the fact that the addition of 2.5 to 5 per cent. of sodium taurocholate to infective blood rendered it incapable of setting up the disease by inoculation.

Heating the virus to 60°C. for an hour was found to destroy its virulence. Exposure to bright sunlight (August) for two hours was also found to effect its destruction. Blood exposed out of doors from December to April, during which time the temperature was often as low as -9°C., was proved to have retained its virulence.

Blood was drawn from 21 horses at periods varying from 3 to 22 months after recovery and injected into healthy animals. All became infected with the exception of two which had received blood from horses which had recovered for three and thirteen months respectively. That these two animals were actually infected, although their blood failed to set up the disease, was proved by the inoculation of healthy horses with materials obtained from various organs when the animals were killed. Further experiments shewed that the blood may retain its virulence for periods ranging up to four years, but that the degree of virulence gradually diminishes.

Experiments were carried out to test the relationship between affected parents and their offspring with regard to the disease.

Blood drawn from four foals, the produce of infected mares, was injected into four healthy horses; one became infected, in two the symptoms produced were doubtful, and in the remaining one the result was negative. The injection of 30 to 100 cc. of milk from infected mares proved successful in three instances, but the milk of infected animals failed to set up the disease by ingestion.

That the progeny of infected mares do not acquire the infection through the medium of the mare's milk was proved by the fact that the intravenous injection of blood from a foal before it had sucked its dam set up the disease.

From this it is concluded that the infection is transmitted hereditarily.

In the donkey the disease generally follows an acute course and death takes place suddenly.

Among species of animals other than the equine the pig is the most susceptible to the virus. In the young goat and sheep a slight febrile condition is produced, but calves, dogs, rabbits, guinea-pigs, and rats are insusceptible.

Details of three experiments with pigs are given.

In the first the subcutaneous inoculation of 10 cc. of virulent blood was followed on the 11th day by a rise of temperature to 40.2°C.

There was interference with locomotion, loss of appetite, acceleration of the heart, frequent shivering fits and, finally, unconsciousness and death.

The second pig inoculated died suddenly during a febrile attack on the fifth day.

The third pig was inoculated with 10 cc. of virulent blood, but failed to shew any evidence of infection. A month later 20 cc. of its blood set up infection in a horse, proving that the virus had been in the pig for that period.

As already mentioned, infection occurs almost entirely while horses are at pasture, very occasional cases occurring in the stable. Experiments were therefore carried out to investigate the following points: Infectivity of the excreta of infected animals; infection through the alimentary canal; infection in the stable; and infection at pasture.

Experiments shewed that the disease could be transmitted by inoculation with infected urine, but no evidence was obtained that the virus was contained in the faeces.

It was also found that the disease could be produced experimentally by the administration of large quantities of infected urine (100 to 200 cc.) by the mouth.

A number of experiments in which 11 healthy horses were stabled with infected animals proved that infection in the stable is of rather rare occurrence. These experiments were carried out during the winter when there were no flies.

Principal importance attaches to the spread of the disease at pasture and in this connection the following points were investigated: The persistence of the virus in pasture during the winter; the manner in which infection occurs when healthy and diseased animals are kept together at pasture; and the relationship of blood-sucking insects to the disease.

In connection with the first of these points experiments shewed that it was quite safe to put healthy horses on to a pasture where numerous cases had occurred during the previous years, but that if healthy horses are pastured with diseased animals or animals that are carriers of the virus there is a very great risk that the healthy animals will become infected. In fact, in the experiments quoted the healthy horses contracted the disease in every instance. The shortest period of incubation was 16 days.

Although the virus is contained in the urine, it requires a large dose of this to produce infection experimentally and, further, the greater part of the urine passed soaks into the ground, that remaining on the top being disinfected by sunlight. Urine can therefore be excluded as the source of infection. This is also indicated by the fact that in stables, where the quantity of excreta is greater than in the open, the disease is far less commonly encountered.

To test the question as to whether blood-sucking insects play any part in the transmission of the disease a number of experiments were carried out.

In two experiments infected horses were kept with healthy ones in insect-proof enclosures for a period of some months. Two out of four healthy animals became infected from one diseased horse kept with them, and in the other experiment one out of two healthy horses

became infected when kept with four animals which were in a very advanced stage of the disease.

This proved that the disease could be transmitted without the agency of insects, provided the place in which the animals were kept was highly infected, but the rate of infection could not be compared with what occurs in the open pasture.

In the second series of experiments healthy and diseased horses were kept separate from each other in fenced areas separated from each other by a distance of about 15 yards. The animals could not mix with each other, but insects could pass freely between the batches.

Three experiments were carried out on these lines. In the first, nine infected horses were kept in one enclosure while four healthy animals were kept in the adjacent one. All the healthy animals contracted the infection.

In the second experiment five infected animals were placed in one enclosure. Two of these died shortly afterwards; one had a severe attack, and in the other two the symptoms were not very marked. Two healthy horses were kept in another enclosure during the whole season without becoming infected, but three out of four healthy animals placed in another enclosure, two at a time, became infected.

The second experiment was repeated the following year with the exception that nine infected animals were used instead of five. In this instance all the healthy animals contracted the disease.

From these experiments the conclusion is drawn that the disease can be transmitted without actual mixing of the diseased and healthy animals, and under conditions which exclude the intervention of insects other than those capable of flying.

Experiments with flies, ticks, etc.

Ticks.—Experiments showed that ticks taken from diseased horses and placed upon healthy horses never transmitted the infection.

Bot-flies.—These are not blood-sucking insects, but to exclude the possibility of the virus being taken into the stomach with the larvae, a number of the latter were taken from the stomach of a diseased horse, crushed in a mortar, and injected into healthy animals. In no instance was the disease transmitted. Further, it is stated that during one year bot-flies were exterminated from all the horses before pasturing, and it was noticed that this had no effect upon the prevalence of the disease.

Mosquitoes can be excluded, as they are found in the stables only, and not on the pastures.

Experiments showed that although sand flies are energetic blood-suckers they are not capable of transmitting the infection.

Negative results were also obtained in experiments in which *Stomoxys calcitrans* was used.

Tabanidae.—These flies are most abundant on the pastures, and energetically attack horses, and particular mention is made of the fact that the spread of the disease coincides with the appearance of this insect. It was found that the flies could not be kept alive for any length of time after they had been caught, and also that they could not be got to suck blood from the animals under experimental conditions.

Although direct proof is not forthcoming, the Committee feel compelled to conclude that the disease is spread through the agency of these flies.

Several species of horse-fly occur on the pastures where the experiments were carried out. The principal ones are the following:—*Chrysops japonicus*, *Haematopota tristis*, *Tabanus chrysurus*, *Tabanus trigonus*, *Tabanus trigeminus*, *Tabanus luridus*, *Tabanus rufidens*, etc.

The lesions present in cases of the disease vary in degree according as the case is acute or chronic, but not in character. They are typically those of anaemia, and in chronic cases there is marked emaciation. The visible mucous membranes are pale, and frequently ecchymosed. The subcutaneous and intramuscular connective tissue contains a gelatinous or blood-stained exudate. The muscles are soft, and pale in colour. The alimentary tract shows no lesions save for catarrh in some cases.

In acute cases, or after several attacks, the spleen may be two or three times the normal size. The capsule may show petechiae, and the pulp presents an appearance resembling that seen in cases of anthrax. In chronic cases the spleen is enlarged and firmer than normal. The capsule of the liver shows white patches of varying size and shape, these being particularly numerous between the lobules.

In acute cases the kidneys are enlarged and softened, and covered with small petechiae.

The lungs do not show any changes peculiar to the disease save oedema, but in some cases there is broncho-pneumonia.

The blood is very watery and greatly reduced in amount. The lymphatic glands, both of the carcass and of the viscera, are enlarged and oedematous, and of a reddish brown colour.

Histological examination reveals changes corresponding to the lesions: Anaemia, extravasations, degenerations of the parenchymatous organs, and the presence of haemoglobin crystals. Special mention is made of an infiltration of small round cells into the parenchymatous organs.

The prominent symptoms of the disease are the irregular occurrence of fever and progressive anaemia. In the majority of cases the fever recurs several times in a month, but sometimes periods of a few weeks up to six months elapse between consecutive febrile attacks.

At the onset of the disease the temperature generally rises suddenly to 39° or 40° C., and after remaining at that level for a day or two suddenly falls again. In chronic cases the temperature remains for some time at a subfebrile level.

During the febrile attacks the pulse rate may be as high as 90 per minute, but it maintains a moderate strength. In the last stages the pulse may be 120 and very weak. Very slight exertion causes an acceleration of the pulse and an increase in the force of the heart beats. Pulsation of the jugulars, which may extend half way up the neck, is frequently seen.

There are little or no disturbances in connection with the lungs, save that exertion causes a distinct acceleration of the respirations, and towards the end froth may be passed through the nostrils owing to oedema of the lungs.

During the febrile attacks there is always albumen in the urine, the

maximum being 10·5 per cent. During remissions and in chronic cases no albumen is found.

In severe or chronic cases the blood clots very slowly, in some cases several days elapsing before the process occurs.

Very few changes are observed in the morphology of the red blood corpuscles. There is a reduction in the number of white corpuscles, but this is not proportional to the decrease observed in the red corpuscles.

All attempts to discover a means of diagnosis apart from clinical methods and the experimental inoculation of susceptible animals failed. A similar result attended all efforts to find a drug which would influence the course of the disease.

Immunisation.—Seven recovered horses were treated with increasing doses of virulent serum and virulent blood in doses of from 2 cc. to 4,000 cc. at intervals of three to six days. Three of these animals survived the treatment, two died of the disease during the course of the immunisation, and in the remaining two cases treatment had to be stopped owing to the development of swellings at the seat of inoculation.

The serum from these immunised horses was found to be of no value as a protective agent, but actually a source of danger, as it was proved to contain the virus of the disease, and to be capable of setting up infection in healthy horses.

It was further found that if the material used as antigen contains blood corpuscles mixed with it a powerful isolysin is produced. Haemolysis occurred not only in infected animals which were treated with the serum, but also in a healthy horse, the count falling from 8,000,000 to 2,700,000 quite suddenly. Virulent serum heated to 60° C. for an hour was proved to be without protective properties.

A number of experiments were carried out with goats, pigs, and calves with the object of ascertaining whether any attenuation could be produced by passing the virus through these animals. No evidence of attenuation could be obtained. In the case of the goat and the calf the virulence appeared to be destroyed.

The addition of 2 per cent. carbolic acid to serum was not sufficient to destroy the virus in less than an hour. Chloroform and toluol failed to destroy it even after remaining in contact with it for 20 hours.

The Report contains a summary of the laws and regulations laid down for the control of the disease.

MISCELLANEOUS.

- 353) LAVERAN (A.) & FRANCHINI (G.). *Infections de Mammifères par des Flagellés d'Invertébrés.* [The Infection of Mammals with the Flagellates of Invertebrates.]—*Bull. Soc. Path. Exot.* 1914. July. Vol. 7. No. 7. pp. 605–612. With 4 text figures.

In previous papers (*Compt. Rend. Acad. Sci.*, 1913, September 1st and November 4th, and 1914, March 16th) the authors have published their observations regarding the infection of mice and white rats with

flagellates derived from the dog flea, the rat flea, *Anopheles maculipennis*, and *Melophagus ovinus*.

Natural infections have been observed to take place in the mouse and rat, and it has been found possible to infect mice by causing them to ingest rat fleas containing flagellates.

Figures are given showing the principal forms in which *Herpetomonas clenoccephali* occurs, and it is said that the flagellates of the rat flea differ but little from those of the dog flea. Rounded or leishmania-forms and flagellate forms occur.

In the case of *Crithidia fasciculata*, the flagellate of *Anopheles maculipennis*, crithidial forms generally predominate but, mixed with these, forms possessing all the characters of the genus *Herpetomonas* are to be found.

The crithidial form also predominates among the flagellates of the intestine of *Melophagus ovinus*.

With whichever of these flagellates the mice or rats are infected the course of the infection is practically the same.

At first the blood is found to contain small oval or rounded intracellular bodies measuring about 2 microns in diameter. The smallest bodies possess only nucleus, but in the larger ones a nucleus and centrosome can be made out. A certain number of these parasites may be observed to be in a state of division.

The "Anaplasma forms" which may be found in the blood of young animals do not appear to be of a parasitic nature.

At a later stage of infection occasional extra-cellular forms may be found. These are fusiform and non-flagellated. They are about five microns in length and possess a nucleus and centrosome.

In a number of cases infection terminates fatally, although parasites are never present in large numbers in these animals. In preparations made from the liver, spleen, and bone marrow free leishmania-like forms predominate. In some preparations from the liver the authors have occasionally observed oval or spherical forms measuring about five microns. In these a nucleus and centrosome can be distinguished, the latter forming the starting point of a flagellum measuring about 12 microns.

Further fresh points are brought forward in the present paper.

Brief details are given of experiments which indicate that it is easy to infect young white mice by allowing them to ingest the faces of rat fleas which contain *Herpetomonas pattoni*.

Endocellular parasites appear in the blood about the third or fourth day, and flagellated parasites a few days later.

Two experiments to infect dogs with the flagellates of the dog flea and the rat flea failed.

A monkey inoculated intraperitoneally with emulsion of the spleen of a mouse infected from rat fleas showed parasites in its blood after about 5 weeks.

In a mouse inoculated with *Crithidia fasciculata* there developed a skin lesion resembling oriental boil, and preparations made from the deep parts of the lesion revealed the presence of leishmania-like bodies. The centrosome of these bodies was rarely bacilliform.

Attempts to cultivate the organisms gave very doubtful results.

The authors conclude that the evidence obtained supports the view that the leishmaniae and the trypanosomes of the vertebrates may have their origin in the flagellates of the invertebrates.

- (354) HOLMES (J. D. E.). *Bursati*.—*Memoirs Dept. Agric. in India*. Veterinary Series. 1914. Sept. Vol. 2. No. 5. pp. 119–153. With 5 plates comprising 11 figures.

This memoir is divided into five parts dealing respectively with the following points: History of Bursati; the Identity of Bursati with Leeches, Summer sore, Granular dermatitis, and other skin affections; Clinical characteristics of Bursati; Histology and Etiology of Bursati; and the Treatment of the condition.

There is also included a list of references and a number of plates, some of which are coloured.

The first part gives some dozen brief references to the opinions expressed by various authors at different times during the last century as to the nature of the disease.

Part II. summarises briefly the views of a number of observers regarding the identity or otherwise of bursati with a number of skin diseases occurring in other parts of the world than India.

Part III. The most accurate description of the clinical features of the disease appears to be that published by SMITH (1874 and 1884). SMITH's account is given at some length and the principal points are the following: The majority of cases occur in the rainy season, but many cases are encountered during the hot weather. The parts generally involved are the angles of the mouth, prepuce, glans penis, fetlocks, and pasterns. The morbid processes start in the subcutaneous tissue. The most distinctive feature of the disease is that it becomes indolent in character, the products of the disease undergoing a peculiar elaboration and remaining as irritants. These products are the so-called "Kunkur stones."

The progress of the formation of a lesion is most easily followed at a place where the skin is thin. There is first of all swelling, and after a few days it is possible to detect that at one point the swelling is becoming harder than elsewhere. This increases in size until it is about the size of a shilling. The skin over the hardened area then becomes thickened and adherent to the tissue beneath. This occurs after about a fortnight. The cuticle is removed either by friction or otherwise and an open sore results. The acute inflammation suddenly disappears and the diseased process becomes an indolent one. The edges of the sore are raised above the surface, are fairly regular and sometimes callous. The ulcerating surface is concave, and situated on and embedded in it are yellowish spherical bodies, which can be pressed out leaving depressions behind. From these depressions a clear liquid exudes. These are the "kunkur bodies."

Although it is common to regard bursati as the result of wound infection, Holmes states that that is not the most common way in which a bursati lesion is started. He is in agreement with SMITH that the lesion is in the first place one of the subcutaneous tissue, and that it is only in the second stage that the skin becomes involved. By the time the skin is lost the underlying tissue has become hard and the surface covered with granulations. In situations where the lesion is not subjected to friction there is little tendency for it to spread, or for the granulations to become very prominent. The surface is dry and sometimes covered with a scab. There may be a

slight serous discharge but, except in old lesions where secondary invasion by pyogenic bacteria has taken place, there is no pus.

Bursati lesions involving mucous membranes are found on the inner canthus of the eye, the angle of the mouth, and the penis. Irregular forms of the disease sometimes occur in which the epidermis is diffusely eroded without any deep-seated lesions being formed.

Holmes has never seen cases of the disease in which there were lesions of the internal organs such as have been described by BURKE, MEYRICK, LINGARD, and others, and he believes that these were cases of mistaken diagnosis.

From the description given it appears that the condition is quite distinct from "leeches," granular dermatitis, summer sores, and other similar skin affections. It is stated that a disease somewhat similar to bursati occurs in America; but there appears to be little doubt that there is some confusion regarding the correct diagnosis of "leeches" and that more than one disease has been described under this name.

In the case of summer sore the calcareous nodules have a very different structure to the kunkurs found in bursati. In summer sore the nodules have an outer covering of fibrous tissue and a central cavity which has a calcified wall.

In bursati the outline of the lesions is not regular, and the kunkur is easily enucleated from the fibrous capsule. The inner mass is hard and fibrous, and is neither caseous nor to any extent calcareous. When pressed between two glass slides it behaves like a hard piece of rubber.

Part IV.—The primary process in the formation of a bursati sore is an area of subcutaneous inflammation due to some specific agent. This irritation, which occurs in a part which has previously had its vitality lowered by some traumatic cause, sets up necrosis of the skin. While this is going on the subcutaneous tissue becomes hard and infiltrated. When the necrotic skin is shed there is left a circular or oval sore with well defined edges. Granulations soon appear in the central part and cause this to become elevated above the surrounding tissues. If such a lesion is kept free from irritation of all kinds it shews little or no tendency to spread, but at the same time no tendency to heal. In debilitated animals, or in cases where the lesion is exposed to irritation, it shews a tendency to spread with the resulting formation of a diffuse tumour in the subcutaneous tissue. The granulations on the surface are very vascular, and pressure causes the expulsion of kunkurs as large as a pin's head or a grain of shot. There is no degeneration of the tissues or pus formation except after invasion with pyogenic bacteria. The deeper fibrous tissue has kunkurs embedded in it and merges gradually into the surrounding tissues.

Microscopical examination of the lesions shews that they are composed of fibrous tissue. In the younger ones there are large numbers of cells of different kinds, and particularly of eosinophiles. In older lesions the fibrous tissue is more dense and giant cells are found. Spaces are found in the sections from which kunkurs have dropped out. These spaces are surrounded by fibrous tissue and in some sections a few pigment-bearing cells may be found. There is no evidence of calcification or other form of degeneration.

The author has been unable to find any evidence that the lesions are caused by filarial embryos, as stated by LINGARD. He has,

however, been able to find a fungoid organism which he believes is responsible for the condition.

The most satisfactory method of examination is as follows: Small blocks of the tumours or kunkurs are washed in alcohol, ether, and water, and then macerated in 40 per cent. caustic potash for 16 to 20 hours. A fragment of such material is taken and pressed between two slides. Isolated spore-like bodies and broken strands of mycelia can be found, but they cannot be satisfactorily differentiated from fragments of cells, granules of eosinophiles and tissue fibres. The most convincing pictures are obtained with material from young lesions. In these can be seen groups of spores which on careful examination are found to be double contoured. The organism is difficult to demonstrate by staining, as it does not take the stain well and is easily decolourised.

Cultivation on Sabouraud's agar most constantly yielded a slow-growing fungus. Growth first became evident on the fifth day in the form of a small dry-looking colony. This gradually spread until the whole surface was covered with a white chalk-like substance which was easily removed, leaving a dry opaque skin adhering to the agar. Cultures of this nature were obtained both from bursati tissue and blood from infected horses.

BUTLER found that the fungus most closely resembled *Sporotricum minutissimum*.

All attempts to infect horses, guinea-pigs, and rabbits failed.

Part V.—The treatment of the condition.—For local treatment the lesions must be completely excised, the wounds being treated as ordinary wounds.

In the experimental treatment of bursati some success has been obtained with arsenic either in conjunction with atoxyl or alone, the treatment being carried out after the manner employed for surra. Under this treatment some cases shewed a rapid improvement, while others did not appear to be benefited.

Some cases shewed rapid recovery when subjected to treatment with biniodide of mercury and potassium iodide, the doses being 5 grains increasing to 20 of the former and 1 to 3 drachms of the latter. Three courses were given of ten to 12 days each with intervals of one week.

HAZELTON recommends that after the diseased tissues have been surgically removed the cavity should be kept constantly dressed with crystalline red oxide of mercury.

Conclusions:

"1. A considerable amount of confusion exists in literature regarding the identity of Bursati in India with 'Leeches' in America, 'Swamp Cancer' in Australia and other affections described as 'Summer Sore,' 'Granular Dermatitis' and 'Parasitic Fibromata,' etc.

"2. The latter affections such as 'Summer Sore,' 'Granular Dermatitis' and 'Parasitic Fibromata,' etc., are held, on the observations of several authors, to be connected with the presence of nematode embryos.

"3. Bursati varies in most clinical aspects from the affections described as 'Summer Sore,' 'Granular Dermatitis' and 'Parasitic Fibromata,' etc.

"4. There is not sufficient evidence to prove that nematode embryos are present in Bursati lesions or that Bursati sores or tumours are caused by such embryos.

"5. There is a similarity in several clinical aspects between 'Leeches' and Bursati.

"The presence of a fungus in 'Leeches' has been recorded by several observers and described as the causal agent. The spores and mycelia of a somewhat similar fungus are frequently found in the kunkur and tumours of Bursati.

"6. The presence of spores and mycelia in the Bursati tumours, and the fact that the cultures of a fungus of the genus *Sporotricum* have repeatedly been obtained from the Bursati tumours and kunkurs and also direct from the blood of horses infected with Bursati, affords some evidence that the disease is a mycosis somewhat resembling the *Sporotricosis* of the horse and mule described by CAROUGEAU in Madagascar."

The paper includes 5 plates comprising 11 figures, shewing the appearance of lesions, spores, and mycelia, and cultures.

(355) HOLMES (J. D. E.) **Some Experiments on the Immunising Effect of the Simultaneous Injection of an Anthrax Attenuated Virus and an Anthrax Anti-serum.**—*Memoirs Dept. Agric. India.* Veterinary Series. 1914. Sept. Vol. 2. No. 7. pp. 199-206.

The experiments recorded in this paper were carried out in order to obtain information regarding the risks attaching to the use of attenuated cultures for the vaccination of animals against anthrax.

Vaccines were prepared by the various known methods, and were tested upon rabbits, guinea-pigs, and sheep, but with none of them was it possible to exclude mortality due to the inoculation. This was especially the case with the sheep. In some cases death from anthrax occurred as late as 20 days after inoculation, the virus apparently regaining its full virulence in the bodies of the animals.

Indian cattle, although susceptible to natural infection by ingestion, are very resistant to experimental inoculation, and therefore cannot be used for testing sera and vaccines.

In the first experiment a number of guinea-pigs, rabbits, sheep, and ponies were inoculated with broth cultures which had been incubated at 37° C. for 48 hours and then heated at 80° C. for one hour. This vaccine was administered in doses of 0.25 to 1 cc. to guinea-pigs and rabbits, 1 to 5 cc. to sheep, and 5 cc. to ponies. Four rabbits were used, and one which received the smaller dose died. Similarly, one guinea-pig out of three used died. Four sheep out of 12 succumbed, but all the three ponies used survived. The unheated culture proved fatal to three sheep in doses of 0.025 cc.

In the second series of experiments a number of sheep were inoculated with 5 cc. of this vaccine and simultaneously with doses of 5 to 25 cc. of a protective serum, which was of such strength that 20 cc. was protective for sheep against inoculation with virulent anthrax. Two of the sheep died in about a week. The surviving five animals were inoculated after eight weeks with 0.025 cc. of virulent culture. All survived.

In the second series of experiments of the same nature 12 sheep received a full dose of protective serum with 1 cc. of vaccine. Two of these died in nine days. One died when the immunity was tested eight weeks later, and one more died when again tested after a further interval of six weeks.

A third series of eight sheep were subjected to a similar experiment, save that the dose of vaccine was 5 cc. All survived the vaccination, but three died within five days when their immunity was tested after an interval of eight weeks.

Tests on ponies.—Eight ponies, ranging from 300 to 400 lb. body weight, were given from 15 to 50 cc. of serum and 5 cc. of vaccine. None died. After eight weeks two succumbed to inoculation with 0.025 cc. of virulent anthrax culture within nine days. Two showed temperature reactions, but lived.

In this experiment one of the three virus control ponies survived.

Five donkeys survived vaccination with 25 cc. of serum and 5 cc. of vaccine, but one died in five days when their immunity was tested with 0.5 cc. of virus after an interval of eight weeks.

Five sheep were inoculated with 5 cc. of vaccine alone. On being tested with 0.025 cc. of virus after an interval of three weeks, all died within three days.

Conclusions :—

"1. The method of immunisation against anthrax by means of the simultaneous injection of serum with the inoculation of an attenuated culture is not free from risk. A varying number of inoculation mortalities follow.

"2. Sero-vaccination confers an active immunity. It does not, however, protect in all cases treated, and the results are variable.

"3. An inoculation of the vaccine alone did not confer any immunity.

"4. The use of an attenuated culture in combination with anti-serum does not appear to be a method in any way superior to the combination of a virulent culture with anti-serum. Following both methods a varying percentage of mortalities from Anthrax occurs, and although an active immunity is produced in the animals which do not succumb, this immunity does not extend to all cases, and a certain number of animals remain susceptible to Anthrax infection."

(356) CUMMINS (S. L.), COPPINGER (C. J.) & URQUHART (A. L.). **Further Observations on the Presence of Antibodies for *Micrococcus melitensis* in the Milk of English Cows.**—*Jl. Royal Army Med. Corps.* 1914. July. Vol. 23. No. 1. pp. 36-41.

In the *Journal of the Royal Army Medical Corps* for January 1914, KENNEDY drew attention to the fact that the sera of five out of 22 English cows examined contained agglutinins for the *Micrococcus melitensis*. The milk of three of these animals gave positive results in high dilutions, and the whey obtained from these milks yielded results exactly similar to those given by the milks. Filtration of the milk or whey appeared to diminish the agglutination. BASSETT-SMITH found one sample of milk out of a number examined which caused agglutination of the organism in dilutions up to 1 in 100. Attempts to cultivate the micrococcus from the milk failed in every case.

These results are of the greatest importance because, if further investigation shows that the reaction is not connected with infection with the organism, the value of the test in the detection of infected goats will have to be reconsidered. If there is found to be a connection between the reaction and infection of English cattle, the question of the infection of the milk will acquire great importance.

The agglutination of the micrococcus by cow's milk has already been observed in Gibraltar, this having been recorded by HORROCKS in 1907. The infection of cows was also referred to in the Report of the Commission on Malta Fever, Part 1, in 1905.

The present paper records the results obtained during the examination of the milk of seven cows, two of which gave positive results.

One of these yielded milk which agglutinated the micrococcus in dilutions varying from 1 in 250 to 1 in 1,000 on different dates.

It was ascertained that a degree of acidity many times higher than that of milk does not cause agglutination of the organism, and that therefore acidity is not a factor in the production of the agglutination.

The milk, whey, and blood serum of the cow all agglutinated the organism to corresponding dilutions. The agglutinin were demonstrable in the milk when tested almost immediately after drawing.

It was also found that heating either the milk, whey or serum to 57° C. for 25 minutes did not destroy the agglutinin, but that the optimum concentration of the agglutinating substance was moved to higher dilutions by heating; thus paradoxal results were obtained. Such paradoxal results were not obtained with unheated milk. This effect was most marked with the blood-serum.

The milk failed to agglutinate *B. typhosus*, *coli*, *dysenteriae*, and *M. paramelitensis*. The whey did not lose its agglutinating power after being kept for three weeks.

In one experiment the passage of diluted whey (1 in 12) through a Doulton filter retarded, but did not eliminate, the agglutinin. The serum of an immune rabbit diluted to the same degree when passed through a filter lost none of its agglutinating power. The rabbit serum when diluted with the whey from the cow and filtered still agglutinated to the same degree, but when whey from a control cow was added complete removal of the agglutinins resulted. These observations are to be repeated, as filters appear to differ in their power of arresting agglutinins.

The milk, whey, and serum of this cow all contained thermo-stable opsonins for the organism. The opsonin was present in large amounts, and was demonstrable in high dilutions. In dilutions up to 1 in 40 the number of cocci per cell were too numerous to allow of accurate enumeration, while with the control serum the number per cell in dilutions from 1 in 5 to 1 in 320 was about 0.5.

Opsonin was still demonstrable after the whey had been kept for three weeks.

The authors have not been able to demonstrate the presence of deviating substances in the heated milk or whey.

In one experiment the heated serum was able completely to deviate two minimum haemolytic doses of complement up to a dilution of 1 in 16. The dilutions were not carried further. In a second experiment no deviation was obtained, but in this instance the serum had been kept diluted with saline for several days, and a positive rabbit serum also yielded negative results when treated in the same way.

The conclusion drawn is:—

The milk, whey, and blood serum of Cow 5 behaves towards *M. melitensis* in a manner comparable to the body-fluids of animals suffering from, or immunised against, this organism, due allowance being made for differences in concentration of "anti-substances" and degrees of immunity.

REPORTS.

(357) HUTCHENS (E.). Annual Report of the Department of Agriculture, Uganda Protectorate. Year Ending March 31st 1914. Report of the Veterinary Department. pp. 28-35. 1914. Kampala. The Uganda Printing and Publishing Company, Ltd.

Rinderpest.—During the year this disease has occurred for the first

time in a district west of the Nile, but up to the time of writing it had been confined to within 25 miles of the river and to the south of the Jinja-Kampala road.

Fifteen cases of the disease were treated by RICHARDSON with permanganate of potash on the lines suggested by WALKER [see this *Bulletin*, Vol. i. No. 2, p. 127.] The results were most hopeful and warrant an extended trial of the method in the treatment of individual cases.

Foot and mouth disease.—The disease is well-known in the Protectorate, but except in neglected cases in transport animals it is not responsible for serious losses.

Trypanosomiasis.—It has been proved by Miss ROBERTSON that *G. morsitans* is the transmitting agent of *T. pecorum* in Uganda. A number of hitherto unrecorded *morsitans* areas have been discovered, among which are Buruli, and two areas in Southern Ankole.

T. nanum was found in a donkey brought down from the Northern Province, and after three months' treatment (method not given), it appeared to have recovered. Trypanosomes of the *T. brucei* type were found in an Egyptian donkey and in a mule. Both animals died, the disease running a rather acute course.

Piroplasmosis in dogs has a wide distribution, but cases met with are seldom fatal if treated with trypanblue.

No cases of rabies have been found.

(358) BEVAN (Ll. E. W.). Report of the Veterinary Bacteriologist, Southern Rhodesia for the Year 1913. fcap. 3 pp.

During the year under review little or no research work has been done, as Bevan was on vacation leave for eight months.

African Coast Fever.—Several outbreaks of this disease occurred. In some instances the circumstances were difficult of explanation. In more than one instance a single case cropped up, generally in a young animal. In other cases outbreaks occurred on old infected areas which according to the accepted theories should have long since been clean. One explanation offered is that an endemic form of the disease allied to the condition known as amakebe in Uganda exists in the Territory. Another is that recovered animals may act as reservoirs of infection.

Anaplasmosis has acquired an increased importance during the year. Not only has it been responsible for a number of deaths among imported animals, but it causes very severe losses in growth and development, which render it one of the greatest handicaps to stock raising. The resistance to the disease possessed by indigenous animals decreases when these are improved by crossing with imported animals, and as the breeding-up extends the disease will acquire more and more importance.

While redwater, the disease with which anaplasmosis is generally associated, is amenable to treatment with trypanblue, anaplasmosis has resisted all methods of treatment tried up to the present.

During the year, 94 animals imported from Great Britain were inoculated, and of these 27 died. In one instance 16 died out of a batch of 33. The deaths took place between the 35th and the 40th day after inoculation. The animal that was the source of the blood

for inoculation was a standard animal which had previously yielded a comparatively mild virus, but on examination its blood was found to contain spirochaetes in addition to anaplasms and piroplasms. It appears to be probable that this infection was acquired naturally through the medium of tick-infested bedding.

A number of experiments were carried out in connection with the immunisation of horses against horse sickness, and the results were considered to be very satisfactory when the animals were tested with very large doses of a certain virus. Unfortunately the animals had to be sold and all trace of many of them was lost, but there is no doubt that under careless management many of them died of the disease. The results, in those cases in which the animals could be followed, indicated that immunisation of an animal against one strain of the virus does not protect against all strains.

A heavy trypanosome infection occurred among trek oxen working in the "fly" area in the Hartley district, but good results were obtained with treatment (method not given). Treatment in pigs has not been successful.

A trypanosome identified by BRUCE as *T. caprae* has been found in the blood of a sheep from Mafungabusi and also in an ox in the Gatooma district.

RECENT LITERATURE.

[Continued from this *Bulletin* Vol. 2, No. 3, pp. 154-156.]

Anaplasmosis and Babesiasis.

- (359) TIBADI (Ettore). Forme di Anaplasma nel Sangue di Diversi Animali in Sardegna. [Types of Anaplasma in the Blood of Various animals in Sardinia].—*Pathologica*. 1914. May 15. Vol. 6. No. 133, pp. 261-264. With 1 text fig.
- (360) VALLADARES (J. F.). Equine Biliary Fever in Madras.—*Parasitology*. 1914. May. Vol. 7. No. 1, pp. 88-94.

Rabies.

- (361) NICOLLE (C.). La Rage en Tunisie pendant l'Année 1913. [Rabies in Tunis during the year 1913].—*Arch. Inst. Past. Tunis*, 1914. Aug. 1. Vol. 9. No. 1, pp. 60-61.

Snake Bite.

- (362) ACTON (H. W.) & KNOWLES (R.). Studies on the Treatment of Snake-Bite. Parts I, II and III.—*Indian Jl. Med. Research*, 1914. July. Vol. 2. No. 1, pp. 46-148. With 5 plates and 3 charts.
- (363) BANNERMAN (W. B.). Report on the Treatment of Snake-Bite with Potassium Permanganate.—*Indian Jl. Med. Research*, 1914. July. Vol. 2. No. 1. pp. 149-182.

Spirochaetosis.

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